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ERRATA

INDIAN JOURNAL OF AGRICULTURAL SCIENCE VOL. V, PART I, FEBRUARY 1935

Page 101, lines 2-5 should read as follows :—

E. A. OSADCHUK. (*Juzno-bereznoe Otdelenie V. I. R'a Gosudarstennyi Nikitskii Botaniceskii Sad*, (*Nikita Botanic Garden*) 1934 : *Bull. No.* **12**, *pp.* 40. (*Full summary of the paper, issued by the Imperial Bureau of Plant Genetics, School of Agriculture, Cambridge, England*).

Page 103, item 10 of table 3, *for* " Persican " *read* " Persican ".

Page 103, item 13 of table 3, *for* " Kizildza Mursal " *read* " Kizildza Mursal "

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ORIGINAL ARTICLES

*AGRICULTURAL METEOROLOGY : STUDIES IN MICRO-CLIMATOLOGY, PART II

BY

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INTRODUCTION

In Part I of this series [Ramdas, Kalamkar and Gadre, 1934] the scope of "micro-climatology" was summarised. This was followed by a brief discussion of different types of instruments for the measurement of air temperature and humidity. Mention was also made of micro-climatic observations taken in the 'open' and inside '*Kharif*' *jowar* and sugarcane during the first half of December 1932 at the Agricultural Meteorological Observatory, Poona.

These observations were taken inside a few crops and in the 'open' daily at the epochs of maximum and minimum temperature. Results of observations during part of the *Rabi* season of the year 1932-33 are discussed in the present paper and an attempt is made to investigate the relationship, if any, between the observations taken inside the crops with those in the 'open'.

The *Rabi* season, October to March or April, is, as mentioned earlier in Part I, a period of clear weather with light winds constituting a climate predominantly of the insolation or radiation type. This season therefore deserves first consideration in any programme of micro-climatic investigations.

We shall discuss here the dry and wet bulb temperatures as well as the pressure of water vapour and percentage humidity calculated from them, with a view to study the modifying influence of different crops on the climate of the open, within the first few feet above ground up to which crops usually grow. While the conditions during individual seasons may vary from year to year it

*The investigations discussed in this series were made in the Agricultural Meteorology Branch (India Meteorological Department), financed by the Imperial Council of Agricultural Research.

may be expected that certain main regularities may become evident during an examination of the data collected even during an individual season.

The role of the humidity factor in micro-climates will be discussed in detail in the next part of this series.

II. Micro-climate of crops and of the open

Micro-climate in the 'open' and inside the following crops are discussed:—

(*a*) *Rabi jowar.*

(*b*) Sugarcane.

(*c*) Wheat.

Details regarding the dates of sowing, variety, seed rate, cultural operations, dates of cropping etc. of the above crops are given in Table I.

TABLE I

Particulars	*Kharif jowar*	Sugarcane	*Rabi jowar*	Wheat
Date of sowing . . .	19th July 1932.	1st Feby. 1932.	7th Oct. 1932.	15th Oct. 1932.
Seed rate per acre . .	8 lbs.	....	7 lbs. (18″ apart).	40 lbs. (1′ apart).
Variety . . .	Gidgap.	Khadya.	Maldandi.	Parner.
Germination . .	25th July 1932.	17th Feby. 1932.	13th Oct. 1932.	20th Oct. 1932.
Plant density per square yard.	12	11 with tillers.	12	126 shoots.
Dates of irrigation (during period under study).	Unirrigated.	Irrigated on 8, 12 December.	Unirrigated.	Unirrigated.
Date of flowering . .	12th Nov. 1932.	30th Nov. 1932.	22nd Dec. 1932.	24th Dec. 1932.
Earthing up . .	....	10th July 1932.	....	....
Date of harvest . .	15th Dec. 1932.	3rd Jan. 1933.	17th Feb.1933.	24th Feb. 1933.
Average height . .	5-8-32—1 to 5″. 20-8-32—6 to 7″. 7-9-32—3′ to 4′. 21-9-32—4′ to 6′. 5-10-32—6′ to 10′. 21-10-32—8′ to 10′.	23-3-32—5″. 8-4-32—6″. 21-4-32—6 5″. 2-5-32—8″. 24-5-32—10″. 4-6-32—15″. 1-7-32—19″. 10-7-32—3′ 7″. 2-12-32—8′ 5″.	1-11-32—6 to 8″. 15-11-32—10 to 16″. 29-11-32—3½ to 4′. 12-12-32—4½ to 5½′. 27-12-32—5 to 6′. 25-1-33—5 to 6′ 9″	27-10-32—6 to 7″. 12-11-32—9 to 10″. 3-12-32—12 to 14″. 15-12-32—15 to 18″ tillers 4-6. 24-12-32—24″. 12-1-33—3′ 6″.

For inter-comparison the temperature and humidity data have been studied at Poona during the following periods:—

(1) Open, sugarcane, *Kharif-jowar*, 4th—15th December 1932.
(2) Open, sugarcane, *Rabi-jowar*, 16th—31st December 1932.
(3) Open, *Rabi jowar*, wheat, 8th—31st January 1933.
(4) Open, *Rabi jowar*, wheat, 1st—21st February 1933.

Some of the periods 1 to 4 could of course be combined. This has not been done in view of the fact that the *Rabi* crops were in a state of growth and it was considered desirable to keep the number of days in each group as small as possible.

The means of the observations of dry bulb, wet bulb, vapour pressure and humidity percentage for the different periods (except the first one for which data have already been given in Part I) are given in Tables II-IV for the minimum and maximum temperature epochs.

The contents of Tables II-IV confirm the conclusions of Part I of this series. The influence of the nature of the crop and its height on micro-climate will be seen from the following pages.

(*a*) *Maximum temperature epoch*

(*1*) *Dry bulb temperature*.—It may be observed from Tables II-IV that at the maximum epoch the dry bulb temperature in the 'open' is very high near the ground and falls rapidly with height. Dry bulb temperatures in *jowar* (an unirrigated crop) near the ground are lower than in the open but at higher levels they approach those of the open and in later stages of growth even exceed slightly the dry bulb temperatures at correspondnig heights in the 'open' (as seen in Tables III and IV). In the case of sugarcane (an irrigated crop) the dry bulb temperatures near the soil are much lower than in *jowar*, and they have a tendency to increase with height; but even at 6 ft. the temperature is about a degree lower than that of the 'open'. The largest difference between the 'open' and sugarcane is about 14°C. near the soil, 3·5°C. at 2 ft. and 2°C. at 4 ft. In the case of the wheat crop (Tables III and IV) the dry bulb temperatures are comparatively higher than that of the 'open' at similar heights; this may be due to the fact that there is ineffective shading of the ground which receives a large amount of insolation and that at the same time air movements or "convective processes" are suppressed by the vegetation.

Table II

Mean dry bulb and wet bulb temperatures, vapour pressure and humidity percentage during the period 16th to 31st December 1932

"Dry Continental Type."

Height	Time	Dry bulb °C.			Wet bulb °C.			Vapour pressure in mm. of mercury			Humidity percentage		
		Open	*Rabi jowar*	Sugar-cane	Open	*Rabi jowar*	Sugar-cane	Open	*Rabi jowar*	Sugar-cane	Open	*Rabi jowar*	Sugar-cane
0·3″	Morning (Minimum Epoch.)	7·7	8·3	9·2	5·2	6·6	8·4	5·4	6·5	7·9	68	78	90
1″		7·3	8·1	8·9	5·0	6·5	8·2	5·4	6·5	7·8	70	81	91
3″		7·1	7·8	8·4	4·9	6·3	7·7	5·5	6·5	7·6	71	81	91
6″		6·9	7·6	8·1	5·0	6·2	7·4	5·6	6·5	7·4	74	81	92
1 ft.		7·1	7·4	7·9	5·3	6·2	7·3	5·9	6·5	7·4	77	84	92
2 ft.		7·4	7·2	7·9	5·8	6·1	7·3	6·2	6·5	7·4	79	85	91
3 ft.		7·8	7·3	8·1	6·3	6·2	7·4	6·5	6·7	7·4	81	86	91
4 ft.		8·3	7·5	8·2	6·9	6·4	7·4	6·8	6·7	7·4	82	86	90
6 ft.		8·7	8·0	8·4	7·2	6·8	7·6	6·9	6·9	7·4	82	84	89
0·3″	Afternoon (Maximum Epoch.)	35·8	30·3	22·4	18·7	18·4	17·2	7·6	9·9	12·2	17	31	60
1″		34·0	30·3	22·8	17·8	17·9	17·1	7·3	9·2	11·8	18	28	56
3″		32·9	29·9	23·3	17·4	17·4	16·8	7·2	8·7	11·3	19	27	53
6″		31·6	29·7	24·0	16·8	17·3	16·6	7·0	8·5	10·5	20	27	47
1 ft.		30·8	29·5	24·8	16·5	16·9	16·2	7·0	8·2	9·5	21	27	41
2 ft.		29·7	29·1	26·2	16·1	16·7	15·7	7·0	8·0	8·2	22	27	32
3 f		29·6	28·9	27·0	16·0	16·5	15·6	6·9	7·9	7·6	22	26	29
4 ft.		29·	28·6	27·2	15·7	16·2	15·5	6·8	7·7	7·4	23	26	27
6 ft.		28·8	28·4	27·7	15·6	16·1	15·5	6·7	7·5	7·1	23	26	26

TABLE III

Mean dry bulb and wet bulb temperatures, vapour pressure and humidity percentage during the period 8th to 31st January 1933

"Dry Continental Type."

Height	Time	Dry bulb °C.			Wet bulb °C.			Vapour pressure in mm. of mercury			Humidity percentage		
		Open	*Rabi jowar*	Wheat	Open	*Rabi jowar*	Wheat	Open	*Rabi jowar*	Wheat	Open	*Rabi jowar*	Wheat
0·3″	Morning (Minimum Epoch.)	9·4	9·7	8·7	6·3	8·0	7·3	5·7	7·3	7·1	63	79	82
1″		9·2	9·6	8·5	6·3	8·0	7·2	5·8	7·4	7·1	65	81	84
3″		8·9	9·4	8·1	6·1	7·9	7·1	5·7	7·3	7·2	65	81	87
6″		8·7	9·3	7·9	6·0	7·8	7·1	5·9	7·3	7·4	68	82	90
1 ft.		8·7	9·1	7·8	6·4	7·7	7·1	6·3	7·4	7·4	72	82	91
2 ft.		9·1	9·1	8·2	6·9	7·7	7·5	6·5	7·4	7·6	73	84	91
3 ft.		9·6	9·1	9·2	7·4	7·8	7·9	6·8	7·4	7·5	74	84	85
4 ft.		9·9	9·2	9·7	7·8	7·8	8·1	7·0	7·4	7·5	75	83	81
6 ft.		10·4	9·9	10·4	8·3	8·3	8·6	7·3	7·5	7·6	76	81	79
0·3″	Afternoon (Maximum Epoch.)	36·2	33·2	36·3	19·5	20·1	20·5	8·7	10·6	10·3	19	27	23
1″		34·2	32·4	35·5	18·3	18·8	20·0	7·9	9·6	9·9	19	26	23
3″		32·9	31·7	34·5	17·9	18·4	19·5	7·9	9·2	9·6	20	26	23
6″		31·8	31·2	33·6	17·3	18·0	18·8	7·7	8·9	8·9	22	26	23
1 ft.		30·9	31·0	32·9	17·0	17·9	18·4	7·7	8·9	8·7	22	26	23
2 ft.		30·1	30·5	31·8	16·7	17·5	17·9	7·7	8·6	8·5	23	26	24
3 ft.		29·6	30·0	30·7	16·5	17·1	17·3	7·6	8·3	8·1	24	26	25
4 ft.		29·2	29·5	30·1	16·4	16·8	16·8	7·6	8·1	7·8	25	26	24
6 ft.		28·7	29·2	29·4	16·1	16·5	16·3	7·5	7·8	7·5	25	25	24

Table IV.

Mean dry bulb and wet bulb temperatures, vapour pressure and humidity percentage during the period 1st to 21st February 1933

"Dry Continental Type."

Height	Time	Dry bulb °C.			Wet bulb °C.			Vapour pressure in mm. of mercury			Humidity percentage		
		Open	*Rabi jowar*	Wheat	Open	*Rabi jowar*	Wheat	Open	*Rabi jowar*	Wheat	Open	*Rabi jowar*	Wheat
0·3″	Morning (Minimum Epoch.)	11·5	11·8	10·4	7·9	9·2	8·4	6·3	7·2	7·3	61	68	77
1″		11·3	11·8	10·2	7·6	9·2	8·2	6·0	7·5	7·3	59	71	77
3″		10·9	11·5	9·9	7·4	9·0	8·2	6·1	7·4	7·3	61	72	79
6″		10·6	11·3	9·8	7·3	8·9	8·2	6·1	7·4	7·5	64	73	81
1 ft.		10·7	11·1	9·8	7·7	8·8	8·2	6·4	7·5	7·4	66	75	81
2 ft.		11·1	11·0	10·2	8·3	8·8	8·5	6·8	7·5	7·5	68	75	79
3 ft.		11·4	11·3	11·1	8·6	9·0	9·0	7·1	7·5	7·6	69	75	76
4 ft.		11·7	11·5	11·7	8·8	9·1	9·3	7·2	7·6	7·6	69	74	73
6 ft.		12·3	12·1	12·3	9·5	9·6	9·7	7·5	7·8	7·8	70	73	72
0·3″	Afternoon (Maximum Epoch.)	39·5	37·7	42·0	19·2	19·5	20·9	6·5	7·9	8·1	12	16	14
1″		37·4	36·4	41·0	18·3	18·7	20·3	6·2	7·0	7·6	13	15	13
3″		36·0	35·5	39·6	17·8	18·0	19·6	6·2	6·7	7·2	13	16	13
6″		34·6	34·7	38·5	17·1	17·6	19·1	5·9	6·5	6·9	15	16	14
1 ft.		33·6	34·0	37·1	16·8	17·3	18·4	5·9	6·4	6·0	15	16	13
2 ft.		33·0	33·1	35·5	16·5	16·7	17·7	6·0	6·1	6·3	15	16	14
3 ft.		32·2	32·6	33·9	16·2	16·7	17·1	5·9	6·3	6·2	16	17	16
4 ft.		32·0	32·1	32·9	16·1	16·3	16·5	5·8	6·0	5·8	16	17	16
6 ft.		31·5	32·0	32·2	15·8	16·1	16·2	5·6	5·8	5·8	16	16	16

(2) *Wet bulb temperature.*—The wet bulb temperatures at the maximum epoch show a decrease with height though less conspicuously than the dry bulb temperatures. The wet bulb temperatures in the 'open' are in general slightly lower than inside *jowar* or wheat (un-irrigated), but in the case of sugarcane (Table II) the values are lower than in the open. Inside wheat (Tables III and IV) the wet bulb temperatures are slightly warmer than inside *jowar.*

(3) *Vapour pressure.*—Vapour pressure inside the crops is higher at all heights compared to the 'open', the highest values being recorded inside sugarcane. There is a tendency for the vapour pressure to decrease with height both in the open and inside the crops.

(4) *Percentage humidity.*—The percentage humidity is minimum near the ground and increases with height in the 'open', the degree of saturation being least in the very warm air layers near the ground.

(*b*) *Minimum temperature epoch*

(1) *Dry bulb temperature.*—As observed in Part I although the variation of micro-climates is less at this epoch, some very interesting features are present. The dry bulb temperatures decrease at first with height both in the 'open' and inside the crops and then begin to increase, the level of inversion being about 6 in. in the 'open', about 1 ft. inside wheat and about 2 ft. inside *jowar* and sugarcane. Temperatures inside tall crops like *jowar* and sugarcane are higher than in the 'open' but tend to the open air value at higher levels. It is remarkable that in the case of the sugarcane field (Table II) which is coolest during the maximum temperature epoch, the minimum temperatures are higher than in the open or inside *jowar.* Inside wheat (a short crop) the minimum temperatures are lower than either in the 'open' or in *jowar.* The explanation of these variations as well as of the coolest air being at some height above ground will be discussed as soon as certain investigations on radiation are completed. They are of special interest in connection with the incidence of frost during winter. Replies to a questionnaire on the frosts which occurred during the last winter lend some support to the generality of the above phenomenon, the problem of frost hazard in India as well as the possible remedial measures to minimise damage are being examined in the light of the various detailed reports received this year. An analysis of the daily minimum temperatures during winter at representative stations in India is also in progress in this connection. Further experimental work at different stations is essential for studying how the frequency of frost varies with height above ground and with locality.

(2) *Wet bulb temperature.*—The wet bulb temperatures at the minimum epoch do not differ very much from the corresponding dry bulb temperatures. The inversions of wet bulb temperature occur more or less at the same height above ground as the latter. The wet bulb temperatures in the 'open' are lower than those inside crops; those in sugarcane are higher than in other crops.

(3) *Vapour pressure.*—Vapour pressure in the 'open' is throughout less than that inside the crops. There is a tendency for the vapour pressure to increase with height in the 'open' and inside *jowar* and wheat. Inside sugarcane, however, it decreases slightly with height in the first foot and is more or less steady at higher levels.

The physical significance of the increase in the vapour pressure with height during night and the reversal of the phenomenon during day has been discussed by Ramdas [1934] and Ramdas and Katti [1934, 1 & 2] in separate papers where the decrease of moisture near the ground at night has been shown to be due to absorption by the soil surface and the reverse effect during day to evaporation from the soil.

(4) *Percentage humidity.*—From Tables II, III and IV it will be noticed that the percentage humidity or the degree of saturation of air with water vapour is lower in the open than inside the crops. In the 'open' as well as inside *jowar* there is a regular increase with height above the ground. In sugarcane (irrigated) the saturation is maximum and more or less uniform at all levels up to 4 feet above which there is a tendency to fall. Inside wheat the percentage is higher than in *jowar* but above 2 feet (the approximate height of the crop) it decreases to the open air values.

III. Correlations between "open" and "crop" temperatures and humidities.

Having seen that the micro-climate of the crops is different from that of the 'open', it is next attempted to investigate whether the micro-climates inside the crops are correlated with conditions in the open.

The values of correlation co-efficients of some of the factors, dry bulb, wet bulb, and vapour pressure for various heights inside the crops with those at corresponding heights in the open are given in Tables V, VI, VII and VIII, both for the minimum and maximum epochs.

Table V

Correlation between temperatures (4th to 31st December 1932)

[For 5 per cent level of significance $r = 0{\cdot}38$]

In the open	With those taken in sugarcane	
	Morning	Afternoon
Surface	·93	·59
1″	·92	·61
3″	·92	·47
6″	·92	·47
1 ft.	·91	·45
2 ft.	·93	·23
3 ft.	·93	·59
4 ft.	·90	·69
6 ft.	·85	·63

TABLE VI

Correlation r between dry and wet bulb temperatures and pressure of water vapour

[For 5 per cent level of significance $r=0{\cdot}58$]

	In the open	With those taken in (4th to 15th December 1932)					
		Dry bulb		Wet bulb		Vapour pressure	
		Jowar	Sugarcane	*Jowar*	Sugarcane	*Jowar*	Sugarcane
Morning.	Surface .	·91	·85	·82	·89	·72	·83
	6″ .	·92	·91	·92	·93	·90	·92
	1 ft. .	·94	·96	·94	·95	.93	·93
	2 ft. .	·94	·98	·90	·97	·89	·95
	4 ft. .	·94	·91	·94	·93	·92	·91
	6 ft. .	·91	·87	·91	·93	·89	·90
Afternoon.	Surface .	·56	·64	·91	·17	·91	·16
	6″ .	·73	·53	·65	·19	·51	·12
	1 ft. .	·59	·50	·60	·56	·78	·33
	2 ft. .	·67	·48	·73	·60	·75	·68
	4 ft. .	·84	·85	·69	·74	·70	·85
	6 ft. .	·82	·78	·86	·81	·85	·91

TABLE VII.

Correlation r between dry and wet bulb temperatures and pressure of water vapour

[For 5 per cent level of significance $r=0{\cdot}40$]

	In the open	With those taken in (8th to 31st January 1933)					
		Dry bulb		Wet bulb		Vapour pressure	
		Jowar	Wheat	*Jowar*	Wheat	*Jowar*	Wheat
Morning.	Surface .	·98	.98	·97	·97	·93	·94
	6″ .	·98	.97	·96	·96	·91	·92
	1 ft. .	·96	.97	·95	·95	·91	·89
	2 ft. .	·97	.97	·96	·95	·92	·89
	4 ft. .	·92	.89	·93	·95	·93	·93
	6 ft. .	·97	.95	·96	·95	·93	·95
Afternoon.	Surface .	·70	·31	·55	·84	·70	·82
	6″ .	·77	·61	·98	·92	·92	·94
	1 ft. .	·70	·59	·75	·80	·97	·94
	2 ft. .	·72	·67	·91	·93	·99	·94
	4 ft. .	·80	·78	·93	·94	·96	·94
	6 ft. .	·84	·83	·95	·97	·97	·97

TABLE VIII

Correlation r between dry bulb and wet bulb temperatures

[For 5 per cent level of significance $r=0{\cdot}43$]

	In the open	With those taken in (1st to 21st February 1933)			
		Dry bulb		Wet bulb	
		Jowar	Wheat	*Jowar*	Wheat
Morning.	Surface	·86	·88	·91	·89
	6″	·91	·91	·94	·91
	1 ft.	·90	·93	·90	·88
	2 ft.	·92	·89	·92	·89
	4 ft.	·90	·91	·95	·95
	6 ft.	·92	·87	·73	·91
Afternoon.	Surface	·36	·05	·79	·58
	6″	·52	·27	·74	·64
	1 ft.	·37	·37	·77	·73
	2 ft.	·46	·54	·64	·65
	4 ft.	·66	·71	·87	·59
	6 ft.	·83	·60	·92	·88

It may be observed that the morning correlations of all the meteorological elements are uniformly high, whereas those for the maximum epoch are smaller especially at lower levels. In the afternoon the correlation improves at higher levels. These results suggest that temperature and humidity inside crops are not related in a simple manner to those in the 'open' and that it may be necessary to measure them directly if one desires to study the actual climatic conditions inside the crops. Further studies on these lines and for different crops are being continued.

Our best thanks are due to the Principal of the Agricultural College, Poona, for giving necessary facilities for this work in the College farm.

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A BIOCHEMICAL STUDY OF THE STARCHES FROM OLD AND NEW GRAIN OF DIFFERENT VARIETIES OF RICE

BY

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I. Introduction

It is supposed that rice of some varieties is fit for being used as boiled rice while that of other varieties is more suited for bread-making than for boiling. There is also a supposition that old rice is more easily digested than new rice. Since rice grain is chiefly made up of starch, and proteids and oil are present only to a very small extent, the differences on account of the varieties and ages must naturally be due mainly to differences in the starchy material. It may be that different varieties of rice contain starches with different reacting powers. It is also quite possible that when rice is stored the starch in the grain undergoes slow change and disintegration, making the whole grain more and more easily digestible with the increase in period of storage.

In order to ascertain whether there are any varietal differences in the appearance and reaction of rice starches and whether during storage the rice starch undergoes any disintegration a regular experimental programme was decided upon. The varieties of rice included in the study were (1) Ambemohor, (2) Kolamba, (3) Patni, (4) Halwar and (5) Mahadi. In studying the differences due to age, rice harvested at the end of 1928 and that at the end of 1930 was taken. The work on 'age difference took some months but all along there was a difference of two years between the old and the new. In addition, there were also some samples of rice one-year, two-year, and four-year old. Description of the varieties of rice used in these experiments are given in a tabular form as under:—

TABLE I

Description of rice varieties

	Ambemohor	Kolamba	Patni	Halwar	Mahadi
Size—					
Length . .	6·6 mm.	7·4 to 8·3 mm.	8·2 mm.	8·5 mm.	9·7 mm.
Breadth .	2·7 mm.	2·4 to 2·5 mm.	3·0 mm.	2·8 mm.	3·8 mm.
Colour of husk .	Brownish to reddish yellow.	Golden to pale yellow.	Pale yellow to whitish.	Pale yellow	Yellow husk and reddish grain.
Grade . .	Medium to fine.	Medium to fine.	Medium coarse.	Medium coarse.	Coarse.
Use . . .	Boiled rice.	Boiled rice.	Boiled rice and bread-making.	Boiled rice and bread-making.	Bread making and cattle feed.
Habitat . .	Maval tract of the Deccan.	North Konkan.	North Konkan.	North Konkan.	North Konkan.

The investigation was carried on along the following lines:—

1. Some physical properties of rice starch such as size of starch grain, action of boiling with water and action of alkali.
2. Digestibility as indicated by hydrolysis with hydrochloric acid, diastase and pancreatin.
3. Liquifaction of starch.
4. Amylohydrolytic enzyme in stored and germinating rice.

When pure rice starch was required for experimental work it was prepared as given below:—Alkali weaker than the strength used did not remove proteinaceous matter sufficiently well while stronger solution attacked the starch grain itself.

The rice was cleaned thoroughly by repeated washings with water. It was steeped in one per cent sodium hydroxide solution and shaken for three hours with the help of hot air engine. The supernatent turbid liquid was removed and the soaked grains were washed with water and ground to a fine condition in a mortar and passed through muslin cloth. The material was treated with

half per cent sodium hydroxide for two days. The alkaline liquid was removed and a small quantity of dilute hydrochloric acid was added to the settled portion to neutralize the alkali. The starch was washed free of chlorides, drained and dried in air. The starch was freed from oil by treatment with ether and alcohol. The purified starch was found to contain 0·013 per cent nitrogen.

II. Some physical properties of rice starch

Size of the rice starch grain

Starch was prepared by the process already described from three varieties—Ambemohor, Halwar and Mahadi, both from the old (1928 harvest) and the new (1930 harvest) samples. The starch was put in water, transferred to a clear slide and examined under a high power with one-twelfth immersion lense. About 200 starch grains of each type were measured with the help of an occular and stage scale. A large number of starch grains from each group gave the following measurements.

Table II

Size of starch grains in mm.

Variety and age	Size in mm.
New Ambemohor	0·0005
Old Ambemohor	0·0005
New Halwar	0·00063
Old Halwar	0·00063
New Mahadi	0·00075
Old Mahadi	0·00075

The new and old samples do not show any difference in size but different varieties do show difference in the size of starch grains. Ambemohor is a fine variety and contains the small-sized grains while Halwar and Mahadi which are coarse and are used for bread-making have comparatively large-sized grains. In general appearance the starch grains of all the varieties are alike and have the usual characteristic polygonal shape.

Behaviour towards methylene blue and iodine

If starches are treated or ground with dye stuffs they exhibit different shades and according to Huberer and Venkatraman [1926 and 1927] it forms a reliable method of distinguishing various starches under the microscope. The rice starches were subjected to these colour tests. A small quantity of starch was made into a thin paste and about two to three drops of half a per cent methylene-blue solution were added to it. After about half an hour a portion of it was transferred to a slide and was viewed under the microscope. No difference in the starch of different varieties or age in rice could be marked. Staining with dilute iodine solution also could not show any difference.

Effects produced by boiling water

Rice grains of different varieties and of different ages, crushed and uncrushed were boiled with water for various periods and observations were made with regard to (1) swelling, (2) softening, (3) breaking of the grain coat and (4) attack on the internal contents. Each of these effects was judged separately by qualitative measurements and these were added and percentage effects were noted. The swelling and softening of the grain are less important than the breaking of the grain-coat and the attack on the internal contents and hence complete swelling and complete softening carried two marks each while complete breaking of the grain-coat and full attack on the internal contents carried three marks each. Thus a complete all-round effect carried ten marks. The following table will show how marks were given and how the total effect was calculated.

TABLE III

Marks under each heading and total marks

Time in minutes	Swelling. Marks out of two	Softening. Marks out of two	Breaking of grain-coat. Marks out of three	Attack on internal contents (Marks out of three)	Total obtained out of 10 obtainable	Total effect
						Per cent
5 minutes .	0·25	0·25	0·25	0·25	1	10
10 minutes .	1	0·5	0·5	0·5	2·5	25

The explanation of the table is as follows:—The rice grains boiled for five minutes got 0 25 marks out of two for swelling, this means the swelling produced was one-eighth of complete swelling. The marks under other items were similarly given. The total of these is one as given in column six. This one is out of ten marks obtainable. The total effect of 5 minutes' boiling is therefore 10 per cent of what the total effect would have been if swelling, softening etc. had all taken place to the fullest extent. The method is arbitrary but some method had to be adopted to give qualitative measurements to show definitely the differences in the effects produced.

The following table gives comparatively the total effect produced by boiling the rice grains of five different varieties for varying periods. In these varieties both the old and the new grains were subjected to the test of boiling with water.

TABLE IV

Total effect produced by boiling rice grains with water for different periods expressed in percentage

Time	Ambemohor		Kolamba		Patni		Halwar		Mahadi	
	New	Old	New	Old	New	Old	New	Old	New	Old
Minutes	Per cent		Per cent		Per cent		Per cent		Per cent	
5 . .	2·5	20	..	20	..	10	0	5	0	2·5
10 . .	5	60	..	60	..	22·5	2·5	8	2·5	3·5
15 . .	30	75	..	75	..	55	7·5	12·5	5	10
30 . .	65	100	..	100	..	65	57·5	57·5	30	50

The effect produced by boiling the rice grains is not equally quick on all the varieties. Ambemohor and Kolamba are the quickest and Mahadi the slowest of the five varieties tried. Halwar is nearly the same as Mahadi with regard to the effects produced. It is these two varieties which show greater resistance to the attack of water than others, and are specially used for bread-making, perhaps because they are not quite so suitable for preparing boiled rice.

The old rice grains are affected more quickly than new rice grains of the same variety, indicating that some change takes place in the grain during storage and it is of such a character that the grain becomes liable to quicker effect of boiling water.

In the case of Ambemohor variety the action of boiling with water was tried on crushed seed also. The action, as expected, is quicker on the crushed than on the uncrushed grains as shown below.

TABLE V

Total effect of boiling with water

Condition	Boiled for 5 minutes	Boiled for 10 minutes	Boiled for 15 minutes	Boiled for 30 minutes
	Per cent	Per cent	Per cent	Per cent
Crushed grains . . .	25	65	80	100
Uncrushed grains . . .	20	60	75	100

Effect of boiling with sodium bicarbonate.—Boiling with half a per cent of sodium bicarbonate solution showed the same differences for the grains of different varieties as boiling with water. In the case of Ambemohor both old and new grains were boiled with half a per cent sodium bicarbonate solution. The comparative figures showing the total effect are given in the following table.

TABLE VI

Total effect of boiling rice grains with half a per cent sodium bicarbonate

Period in minutes	Ambemohor		Kolamba	Patni	Halwar	Mahadi
	New	Old	Old	Old	Old	Old
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
5 . . .	2·5	35	35	10	7·5	5
10 . . .	7·5	65	65	30	15	12·5
15 . . .	35	80	80	60	50	20
30 . . .	75	100	100	80	65	60

Action of caustic soda on the rice grains.—Warth and Darabsett [1914] studied the disintegration of rice grains with the object of classifying the different varieties of rice according to the resistance they offered to the alkali

treatment. They came to the conclusion that on this basis rices could be classified. The disintegration roughly corresponded with the quality of the starch. Rice grains which contained much of liquifiable starch, disintegrated easily. The coarse variety grains were not so easily attacked as the grains of the finer varieties. Warth and Darabsett studied the disintegration with the help of microphotographs of the attacked grains and also from the general breaking down of the grains. For the purpose of this paper the comparative general breaking down of the grains was studied. It included observations on the following:—(1) swelling of the grain, (2) decolourisation, (3) transparency, (4) breaking of the seed-coat, (5) softening of the grain and (6) effect on the internal contents. Marks were assigned for comparison. Complete action in each of the first three items carried one mark, complete action in each of the next two items carried two marks and the last item carried three marks for its complete action. The total marks for all the complete actions were ten and the percentage of the total of all the effects in each case was calculated as explained under the effects produced by boiling with water.

The first effect observed of the treatment with alkali, is the decolourisation of the grains and softening them. There is a general swelling followed by the breaking down of the coat of grain. Ultimately the starch is attacked which becomes gelatinised and transparent.

The strengths of the caustic soda used were (1) two per cent, (2) three per cent, (3) four per cent and (4) five per cent, and the periods for which the grains were subjected to the action of caustic soda were (1) one hour, (2) two hours, (3) three hours, (4) four hours and (5) twenty-four hours. Ten grains (two years old) were studied under each treatment the quantity of the alkali being 10 c.c. for every strength.

The table gives the total effect produced on rice grains by treatment with different strengths of caustic soda.

TABLE VII

Total effect on rice grains produced by caustic soda treatments expressed in per cent

Variety	Period of treatment				
	One hour	Two hours	Three hours	Four hours	Twenty-four hours
			Two per cent caustic soda solution		
Ambemohor	10	12	17	60	..
Kolamba	10	12	17	60	..
Patni	7	7	7	30	..
Halwar	7	7	7	17	..
Mahadi	5	5	7	15	..
			Three per cent caustic soda solution		
Ambemohor	15	20	60	70	97
Kolamba	15	20	60	70	97
Patni	12	15	60	70	97
Halwar	10	12	17	25	95
Mahadi	10	10	15	22	80
			Four per cent caustic soda solution		
Ambemohor	55	82	100	100	100
Kolamba	55	82	100	100	100
Patni	12	32	55	70	100
Halwar	7	25	45	65	92
Mahadi	5	15	35	37	85
			Five per cent caustic soda solution		
Ambemohor	60	85	100	100	100
Kolamba	60	85	100	100	100
Patni	15	62	70	80	100
Halwar	7	42	47	70	92
Mahadi	7	30	40	62	87

Caustic soda acts on rice grain breaking the coat, softening the internal contents and gelatinising the starch, its action increases with the strength of the solution and with the length of the period for which the caustic soda solution acts on the grain. All the varieties are not equally attacked. There is a varietal difference shown in the resistance offered by the grains to the attack of the alkali. Ambemohor and Kolamba are the easiest attacked out of the five varieties tried and the last two, namely the Halwar and the Mahadi, are the most resistant. This again shows that it is the fine varieties which show less resistance in breaking down or in disintegration than the coarse varieties used ordinarily for bread-making. Ambemohor breaks down to the extent of 60 per cent in four hours even with such a weak solution as two per cent caustic soda while the Mahadi variety requires five per cent solution to bring about that effect in the same time.

The action of caustic soda was used to see if rice grains of different ages showed difference in resistance. Rice grains of three varieties were tried with three per cent solution for varying periods. The results show that the older the grain the less is its resistance to the attack of caustic soda. This is shown by the figures in the following table.

TABLE VIII

Total effect on new and old grains by the action of three per cent caustic soda expressed in per cent

Period	Ambemohor		Halwar		Mahadi	
	New	Old	New	Old	New	Old
Hour						
1 . . .	10	15	5	10	5	10
2 . . .	12	20	5	12	5	10
3 . . .	40	60	12	17	12	15
4 . . .	60	70	17	25	15	22
24 . . .	95	97	85	90	70	80

Rice grains of the Ambemohor variety were subjected to the treatment of three per cent caustic soda at two different temperatures—27°C. and 37°C.—under crushed and uncrushed conditions and also under polished and unpolished conditions. Difference in condition always gave difference in the resistance of the grain to the attack of the alkali. Higher temperature produced quicker reaction than lower temperature. Unpolished grains showed greater resistance than polished grains and uncrushed grains showed greater resistance than crushed grains.

Conclusions

The following conclusions can be drawn from the results of the experiments given in this section. The results, it must be admitted, are worked out after the fashion of score-card judging. They are intended to give a comparative idea about the disintegration of rice grains under different conditions.

(1) Boiling with water affects the rice grain. The longer the boiling the greater is the effect.

(2) The effect of boiling with water is greater on old rice grains than on new grains.

(3) Boiling with water does not affect the rice grains of all varieties equally. Those that are generally used for bread-making show greater resistance than those used for boiled rice.

(4) Crushed grains are naturally attacked more quickly than uncrushed ones.

Similar conclusions can be drawn from the experiments done with caustic soda treatment.

(5) Varietal and age differences are shown by treating rice grains with caustic soda. The caustic soda attacks grains of all the varieties and ages.

(6) The longer the period of attack and the higher the strength of the caustic soda solution the greater is the effect produced.

(7) Rice varieties for bread-making are more resistant than fine varieties used for boiled rice.

(8) New rice grains show greater resistance than old grains.

(9) Crushed and polished condition as also higher temperature tend to increase the effect on the grain.

III. Hydrolysis of rice starch with hydrochloric acid and diastase and pancreatin

As the chief object of the investigation was to study the differences in the digestibility of rice of different varieties and different ages, experiments were arranged to see the effects of hydrochloric acid on starches and flours, and of diastase and pancreatin on flours. Many workers have studied the hydrolysing action of hydrochloric acid and enzymes on starches of various food grains. Stone [1896] carried on experiments to study "the degree of susceptibility of different starches to the action of different enzymes". He established an order of decreasing susceptibility of various starches. He also mentions Lenberg [1877] and Hammersten [1883]. Each one of these gave his own order of susceptibility for saccharification. Rockwood [1910] studied the action of pancreatin, ptylin and diastase on the starch from bleached and unbleached wheat flours and he found that the reducing sugars were formed at equal speed in both the cases as tested by starch-iodine reaction. Langworthy and Deuel [1920] carried on interesting work on the digestibility of certain starches on human subjects. A good deal of work has been done by several workers like Brown and Morris [1895], Ling and Baker [1895], Lintner [1895] and others.

Action of hydrochloric acid on starches

By way of trial hydrochloric acid of 2·5, 25, 50, 75, and 100 per cent was used but it was observed that 100 and 75 per cent hydrochloric acid was too quick in its action to show differences in its actions on different starches while any strength lower than 25 per cent proved to be too slow. Fifty per cent strength was found to be the most suitable for measuring differences and hence that strength was used. Starches from three different varieties and of different ages were experimented upon.

In each case one grm. of starch was made into a paste with 5 c.c. of water. 25 c. c. of 50 per cent hydrochloric acid was then added and the solution thoroughly stirred to get a uniform mixture. The hydrolysis was allowed to proceed for a fixed period at a temperature of 27°C. At the end of the period the acid was neutralised with sodium carbonate and the whole volume made up to 200 c. c. The solution was then filtered through dry filter paper and titrated against Benedict's [1910-11] solution which was standardised. The reducing sugars were estimated as dextrose and the figures for dextrose were multiplied by 0·93 (conversion factor for starch) to get starch. Thus the amount of starch digested was obtained from which the per cent digested was calculated. Each set had a duplicate. The results of the duplicates agreed with each other and hence there was no need to take averages. The hydrolysis was carried on in one set for 24 hours and in another for 72 hours. The varieties used were (1) Ambemohor, (2) Halwar and (3) Mahadi, both new and two years old.

TABLE IX

Percentage of starch hydrolysed

Variety	Age	Hydrolysis	
		24 hours	72 hours
		per cent	per cent
Ambemohor	New . .	34·29	64·62
	2 year old .	46·80	76·14
Halwar	New . .	33·84	63·90
	2 year old .	46·80	76·14
Mahadi	New . .	34·29	63·45
	2 year old .	45·18	74·97

The figures show that the longer the period of attack the greater is the amount of hydrolysis. Both for 24 hours and 72 hours digestion the old starches are more easily attacked than the new ones. There is, however, very little difference shown by different varieties with regard to the hydrolysis by 50 per cent hydrochloric acid.

Action of hydrochloric acid on rice flour

It is after all the rice flour and not the purified starch that is used as food and hence the hydrolysing action of hydrochloric acid, 50 per cent. in strength, was tried on flours. Rices of the same three varieties and the same two different ages were used. The polished rice grains of each type were ground to a fine powder. Half a grm. of the flour was made into a paste with 10 c.c. water and to this 10 c.c. of 50 per cent. hydrochloric acid was added and well stirred. The hydrolysis was carried on at a temperature of 27°C. for 48 hours. The acid was neutralised with sodium carbonate and the estimation of reducing sugars and calculation of digested starch was done as in the previous experiments.

The results obtained are given in the following table. They are of hydrolysed starch per 100 grms. of the flour and not per 100 grms. of starch in the flour.

TABLE X

Per cent of hydrolysed starch with 10 c.c. of 50 per cent hydrochloric acid at 27°C. in 48 hours

Variety	Year of harvest	Starch hydrolysed
		per cent
Ambemohor		
New	1930	7·74
Two year old	1928	8·46
Halwar		
New	1930	6·30
Two year old	1928	6·66
Mahadi		
New	1930	5·76
Two year old	1928	7·20

The flours from two year old rices are more easily attacked than those of new rices.

The same rice samples (those of 1928 harvest and 1930 harvest) were stored for a period of two years more, that is to the end of 1932 and were subjected to the action of 50 per cent. hydrochloric acid exactly in the same manner as in the previous experiments.

Rice grains harvested at the end of 1928 were four years old and those harvested at the end of 1930 were two years old at the time they were experimented upon. The above table gives the difference between new and two-year old rices while the following table gives the difference between two-year old and four-year old rices.

TABLE XI

Per cent of hydrolysed starch by 10 c.c. of 50 per cent hydrochloric acid at 27°C. in 48 hours

Variety	Age	Harvested	Starch hydrolysed
		Year	Per cent
Ambemohor	2 year old .	1930	12·82
	4 year old .	1928	14·18
Halwar	2 year old .	1930	9·86
	4 year old .	1928	10·52
Mahadi	2 year old .	1930	9·10
	4 year old .	1928	10·14

Flour from four-year old rice is more easily broken down than the flour from two-year old rice. These, however, were samples from different stocks and to get a better idea, samples from the same stock should be compared. Taking the same sample of 1928 harvest kept for two years and hydrolysed in 1930 and for four years and hydrolysed in 1932 we can put the figures as follows:—

TABLE XII

Per cent of hydrolysed starch by 10 c.c. of 50 per cent hydrochloric acid at 27°C. in 48 hours

Variety	The same stock	
	Two year old (Harvested in 1928 and hydrolysed in 1930)	Four year old (Harvested in 1928 and hydrolysed in 1932)
	Per cent	Per cent
Ambemohor	8·46	14·18
Halwar	6·66	10·52
Mahadi	7·20	10·14

These figures show clearly the change produced during a period of two years in storage; with increasing age the rice becomes easier for attack by chemical and other agencies.

Action of diastase absolute

Diastase is an enzyme which acts on starch breaking it down to reducing sugars. It is, therefore, a good reagent to measure the differences between varieties and ages of rice. The strength of diastase used was 0·1 per cent of Merck's diastase absolute. The action was tried for one-hour-and-a-half at a constant temperature of 37°C. The object in selecting 37°C. temperature was to try the digestion of starches by the enzyme at body temperature. Blank experiment with diastase only was carried on to ascertain if any quantity of reducing sugars was produced by the enzyme and to make necessary correction in the results obtained, but the diastase used did not give any reducing sugars. The strength of the diastase activity was compared by experimental trials and it was found that one c.c. of one per cent diastase was equivalent to 0·75 c.c. of concentrated hydrochloric acid (sp. gr. 1·16) under the same condition of temperature, dilution, etc.

Half a grm. of the flour was made into a thin paste with 10 c.c. water in a large test-tube and was gelatinised by keeping it in a boiling water-bath for three minutes. It was then cooled to room temperature and 100 c.c. of diastase solution of required strength was added to the gelatinised starch. To prevent fermentative and other changes a few drops of toluene were added to

the mixture. The tube was after that immediately put into a water-bath kept at a constant temperature of 37°C. The reaction was allowed to continue for the required period. The enzyme was killed by adding a few drops of 10 per cent caustic soda and by immersing the test tube in boiling water for about 10 minutes. The contents of the tube were cooled to room temperature and made to a known volume in a graduated flask. The solution was filtered through dry filter paper and titrated against standardised Benedict's solution for the estimation of reducing sugars formed. The figure for reducing sugars was multiplied by 0·93 to get the amount of starch and the percentage of starch hydrolysed was calculated. A number of such tubes necessary for the experiment were handled in one lot. Each trial had a duplicate and the duplicates agreed with each other.

As in the case of hydrochloric acid three varieties of rice—Ambemohor, Halwar and Mahadi, both new and two-year old were tried, and the results obtained with 0·1 per cent diastase solution for one hour-and-a-half at 37°C. are given below.

TABLE XIII

Per cent of hydrolysed starch with 10 c.c. of 0·1 per cent diastase solution at 37° C. for 1·5 hours

Variety	Year of harvest	Starch hydrolysed
	End of	Per cent
Ambemohor		
New	1930	21·60
Two year old	1928	31·68
Halwar		
New	1930	8·70
Two year old	1928	16·12
Mahadi		
New	1930	18·00
Two year old	1928	31·14

The results also indicate that there are differences in the starches of different varieties with regard to the action of diastase. Flours of old grain are attacked more easily than those of new grains.

The same rice samples were stored for a period of two years more and were then subjected to the action of diastase as above to get the difference between two-year old and four-year old rices.

TABLE XIV

Per cent of hydrolysed starch with 10 c.c. of 0·1 per cent diastase solution at 37° C for 1·5 hours

Variety	Age	Harvest	Starch hydrolysed
			Per cent
Ambemohor	2 year old .	1930	24·70
	4 year old .	1928	35·83
Halwar	2 year old .	1930	11·90
	4 year old .	1928	17·16
Mahadi	2 year old .	1930	18·80
	4 year old .	1928	35·40

The two-year old and four-year old samples show a distinct difference. The samples of two-year old and four-year old as compared in the above table do not belong to the same stock. Figures from Tables XIII and XIV can be taken and arranged as below to show the difference between the two-year old and four-year old rices belonging to the same original stock for proper comparison.

TABLE XV

Per cent of hydrolysed starch with 10 c.c. 0·1 per cent diastase solution at 37° C. for 1·5 hours

Variety	Two year old	Four year old
	Per cent	Per cent
Ambemohor	31·68	35·83
Halwar	16·12	17·16
Mahadi	31·14	35·40

This clearly indicates the change produced during a period of two years in storage. In the above experiments samples from one and the same stock were taken and hydrolysed at various times to see the effect produced during storage. Another experiment was arranged in which rices belonging to the same variety and strain but harvested in different years were subjected to the action of hydrolysis at one and the same time. For this purpose only one variety was taken and that was Ambemohor. Grains from the harvests of 1928, 1930, 1931 and 1932 were taken and subjected to hydrolysis in the beginning of 1933 so that there was a new sample collected at the end of 1932, one-year old sample, two-year old sample and four-year old sample. Only one period of reaction was tried. The material consisted of finely ground rices of different ages. 20 c.c. of concentrated hydrochloric acid was used with one set and 20 c.c. of one per cent diastase solution was used with another set. Period of action was 24 hours kept at 27°C. The estimations were made as in previous calculations, the percentages being determined on flours.

TABLE XVI

Per cent of hydrolysed starch in 24 hours at 27° C.

Year of harvest	Age	With concentrated hydrochloric acid	With one per cent diastase
		Per cent	Per cent
1928	Four year old	58·99	49·40
1930	Two year old	50·12	45·45
1931	One year old	47·61	43·64
1932	New	46·50	41·17

The new rice shows the least hydrolysis. As the age of the grain increases the percentage of hydrolysis also increases. The results obtained with hydrochloric acid and with diastase are quite parallel.

Action of pancreatin

It has been shown that there is a clear difference between starches from old and new samples of rice when they are acted upon by hydrochloric acid and by diastase. Pancreatin is another enzyme which acts on the starch and hence the action of this also was tried to see if it could distinguish between the old and the new rices. The same varieties and the same ages of rices were done in the same manner as in the previous experiments.

One-fourth grm. of starch was taken and was gelatinised in test tubes with 10 c.c. water. After cooling to the room temperature 10 c.c. of different strengths of pancreatin were added and the reaction was allowed to proceed for a definite period at 37°C. A few drops of toluene were added to prevent bacterial action. It was observed that pancreatin was not easily soluble in water and hence it was shaken vigorously for a minute and a measured portion was pipetted out as required. After the digestion period was over the test-tubes with starch and pancreatin were put in a boiling water-bath for 15 minutes to kill the pancreatin. The solutions were made to a known volume and filtered through dry filter paper. After titration against Benedict's standardised solution the dextrose figures were multiplied by 0·93 to get starch. The percentage figures were then calculated.

The strengths of pancreatin taken were 10 c.c. of 0·5 per cent and 10 c.c. of one per cent and the periods of digestion were two hours and three hours. In each case duplicates were done and they agreed with each other.

TABLE XVII

Per cent of starch hydrolysed with pancreatin

Variety	Half per cent pancreatin		One per cent pancreatin	
	Two hours	Three hours	Two hours	Three hours
	Per cent	Per cent	Per cent	Per cent
Ambemohor				
New	Traces	Traces	4·32	4·93
2 year old	29·52	36·00	32·24	43·36
Halwar				
New	13·68	14·76	20·16	32·24
2 year old	24·12	26.28	30·60	44·64
Mahadi				
New	25·92	36·00	34·56	41·76
2 year old	33·12	45·56	44·28	49·68

It will be seen that there is a distinct difference in the susceptibility of the starches of new and old rices towards pancreatin. The starch prepared from old rices undergoes greater amount of hydrolysis than that prepared from new ones. The effect of prolonging the period of reaction or increasing the strength of the enzyme is to increase the amount of action. In all cases the difference between the old and the new rices persists. One striking feature in the above experiments is that the starch from new Ambemohor is practically not acted upon by 0·5 per cent pancreatin and only slightly acted upon, as compared to other starches, by one per cent pancreatin. In the previous experiments Ambemohor starch was the quickest to be acted upon by hydrochloric acid and also by diastase. With pancreation, however, that position does not remain the same Especially new Ambemohor seems to resist the action of pancreatin very much.

Conclusions

Rices of three varieties—Ambemohor, Halwar and Mahadi—both old and new were subjected to the hydrolysing action of hydrochloric acid, diastase and pancreatin to ascertain if the amount of action produced showed any difference in the old and new rices.

The hydrolysing action of different strengths of hydrochloric acid, diastase and pancreatin acting for varying periods was more on old rices than on new ones whether used as flour or as prepared starch.

If the three rices are hydrolysed at one time and again after two years a distinct difference in the percentage of hydrolysis is seen, the second trial (after two years) giving greater hydrolysis than the first. This indicates that as rices grow old they become more susceptible to the action of hydrolysis.

If the rices of the same strain and variety but harvested in different years are subjected to hydrolysis, a distinct difference is seen. The oldest rices are attacked the most and the new ones the least.

With hydrochloric acid and diastase Ambemohor starch is hydrolysed more quickly than that of the other varieties used for bread-making. This, however, does not hold good with pancreatin.

In all cases the hydrolysis increased with increasing strength of the reagents and with increase in the period of action.

All the reactions, therefore, point to the conclusion that old rices are easier to digest than the new ones.

IV. Liquefaetion of starch in new and old rices

Introductory

The general belief that old rices are more easily digested than the new ones has been confirmed by the results obtained so far both qualitatively and quantitatively. Now the question arises as to why there is such difference in the behaviour of the two. There are two probable answers to this question, one of which is that the starch in the old rices may break down into more easily digestible portions during the resting period, and the other is that during the

process of cooking the starch of the old rices may undergo greater liquefaction than that of the new ones, so that it is better attacked by the digestive enzymes of the human system.

As regards the first possibility it has been found out that the resting seeds do contain an Amylo-hydrolytic-enzyme (proved further on) and if this is active during the resting period, it may attack the starch of the grains and thus turn it into simpler units. This enzymic breaking-down may be further helped by a physical disintegration of the grain, and these two processes may make the old rices more susceptible to the action of the digestive ferments. This section, however, aims at studying the liquefaction of starch at different temperatures, with the object of finding out differences in the starch prepared from new and old rices if there be any, and thus to account for the difference in the digestibility of the two to a certain extent. That the cooking quality of starch is closely co-related with the extent of its liquefaction has long been shown by the work of Warth and Darabsett [1914] and needs no further explanation.

It is however interesting to note at this stage what happens to the starch during the process of cooking. The most important change that takes place is that the structure of the grain changes to a great extent both internally and externally. The granules break down and rush out of the ruptured cell-walls. This gives the characteristic jelly-like appearance to the whole swollen mass, when probably the starch in the grain is said to be gelatinised. In the process of gelatinisation the starch is to a great extent liquefied and it is this liquefied starch that is most easily attacked by the digestive enzymes.

Fofanow [1911] carried on some experiments on human subjects with raw starch of wheat, oat, rice, potato and he came to the conclusion that all of them were completely assimilated but that potato starch was less digestible than others. He further said in his work that "numerous artificial experiments have proved that the raw starches are more slowly attacked than those that have been cooked, by the digestive ferments".

Langworthy and Deuel [1920] who studied the digestion of raw wheat, corn, and potato starches came to the conclusion that all of them were completely assimilated though of course the potato starch caused some physiological disturbance. They further say that "Diastase has only a feeble action on uncooked starch".

From the work of the above workers it may be seen how cooking facilitates the digestion of starch and what role it plays in the chemical and physical changes that the starch undergoes during this process. The only point with which we are concerned is the liquefaction of the starch during the process of cooking.

Experimental

Two grms. of rice in a finely powdered condition was mixed with 25 c.c. of distilled water and was kept at fixed temperatures for one hour and a half, the temperatures being regulated in a water-bath. Thus all the starch that could be liquefied at that particular temperature was present in the solution. The contents of the test-tubes were then hydrolysed at 27°C. by 5 c.c. of one per cent diastase absolute, for two hours. The solution was then made to a known

volume after killing the enzyme with two to three drops of strong caustic soda. The solution was then filtered and 50 c.c. of it was subsequently hydrolysed by 15 c.c. of hydrochloric acid by keeping the containers in a boiling water-bath for two hours. The contents were further made to a known volume, after neutralising the excess of the acid with sodium carbonate, and the reducing sugars were estimated as dextrose by titration against standardised Benedict's solution. From this the amount of starch liquefied at that particular temperature was calculated. Only three varieties of rices were studied and these belonged to 1930 and 1928 harvests.

The following temperatures were tried for the liquefaction:—(1) 55°C. (2) 65°C. (3) 75°C. and (4) 95°C.

Commercial diastase does not act on crude starch. This means that in the present trials it is only the liquefied starch that is attacked by the commercial diastase used, at the different temperatures tried. The following are the results obtained in the present trials.

TABLE XVIII

Per cent. liquefaction of starch in the new and old rices

Variety	Liquefaction			
	55°C.	65°C.	75°C.	95°C.
	Per cent	Per cent	Per cent	Per cent
New Ambemohor	7·06	12·38	29·76	46·50
Old Ambemohor	7·83	15·49	37·20	49·46
New Patni	6·88	14·88	25·66	45·08
Old Patni	7·44	21·24	29·74	58·12
New Mahadi	6·88	10·04	20·27	40·21
Old Mahadi	8·74	11·34	24·74	49·46

The figures of the above table show that (1) the percentage liquefaction in general increases along with the temperature and is higher with higher temperature. (2) There is a significant difference in the behaviour of new and old rices so far as the liquefaction of the starch in them is concerned. The starch in the old rices undergoes greater amount of liquefaction than that in the new ones. This is perhaps the reason why old rices are more easily digested than the new ones.

V. Study of the amylase enzyme developed in the resting and germinating seede of rice

Introductory

It has been noted that during the process of "keeping" the rice seeds undergo a sort of disintegration and that both the grain and the starch are comparatively easily acted upon by diastase, pancreatin and hydrochloric acid. It is to be seen whether this breaking down, and easy susceptibility to the action of hydrolysing agents is an enzymic process or is due to any physical disintegration of both the grain and the starch during the course of storing time. With this object it was decided to study the amylase enzymes that are developed in the resting and germinating seeds of rice with a view to find out the difference, if there is any, in the behaviour of the two and also to compare the activities of the two with that of standardised solution of diastase absolute (obtained from E. Merck), under the same conditions of temperature and dilution.

Brown and Morris [1895] are of opinion that the "Saccharifying ferment" is entirely a product of germination and that ungerminated barley does not contain any starch-transforming body. In the same paper however they quote Kjeldahl's opinion on this point (Resume De Comptes Rendus—1875—I—129) which runs as follows:—

"He has shown the existence of an active amylohydrolytic enzyme in ungerminated barley and though this has a considerable saccharifying power upon soluble starch yet it has scarcely any influence in liquefying starch-paste." The enzyme of the resting seeds is thus differentiated from that of the germinated seeds in not having the power of liquefying starch.

It will thus be seen that an amount of work of a similar nature has been done in the past, though not on the rice seed. The following experiments were arranged to see whether amylohydrolytic action goes on very slowly in the resting seed making the seed more and more susceptible to the attack when favourable conditions occur. In this connection the amount of reducing sugars formed by the action of the enzyme alone has been taken as a measure of the activity of the enzyme. Trials with germinating seed also were taken for comparison.

Experimental

For the study of the enzymes of the resting and germinating rice seeds, the experiment was laid out into nine different lots and each lot was analysed separately for dextrose, the amount and formation of which was taken as a measure of the activity of the enzyme. In the case of all the lots duplicate trials were made and the dextrose formed as a result of the action of the enzyme was estimated by Benedict's method. In the case of the resting seeds those of previous year's harvest were used and in the case of germinated seeds those that were in the soil for 96 hours (4 days) were used. All natural conditions regarding the amount of moisture, uniform sowing, etc. were given to the seeds and the germination was complete by the 4th day, when the seeds had put forward

both the radicle and the plumule. The average length of the plumule was about 1·25 cm. and that of the radicle was nearly one cm. The plumule was slightly green in colour.

Seeds of the above description were used in the lot of "germinated seeds" and before preparing an extract with distilled water, they were completely freed of all the adhering soil and the portions of shoots and roots. The following were the lots made in the present experiment:—

Lot I.—Blank—*i.e.* Extract of resting seeds.

Lot II.—Blank—*i.e.* Extract of germinated seeds.

Lot III.—Blank—*i.e.* Extract of 0·5 per cent. diastase absolute.

Lot IV.—Ungelatinised starch *plus* extract of resting seeds.

Lot V.—Ungelatinised starch *plus* extract of germinated seeds.

Lot VI.—Ungelatinised starch *plus* extract of 0·5 per cent diastase absolute.

Lot VII.—Gelatinised starch *plus* extract of resting seeds.

Lot VIII.—Gelatinised starch *plus* extract of germinated seeds.

Lot IX.—Gelatinised starch *plus* extract of 0·5 per cent diastase absolute.

Preparation of the extracts

In the case of resting seeds those of two-year old harvest were used, and in the case of germinated seeds one-year old seeds which were in the soil for 4 days were used. The germinated seeds were cleaned of all their adhering dirt and the portions of the shoots and roots, and then the extract of the same was made. In both the lots 30 grms. of seeds (germinated and resting) were used and they were extracted with 100 c.c. of distilled water for 24 hours, after shaking the same on a machine run by hot air, for 5 hours. During extraction a few c.c. of toluene were added to prevent fermentation and other changes. These extracts were used for the hydrolysis of the rice starch. As said before the experiment was laid out into 9 different lots and the hydrolysis was carried on at the temperature of 27°C. under the same conditions of dilution, etc. In the case of diastase solution 20 c.c. of 0·5 per cent strength was used and in the case of all other extracts the same volume *i.e.* 20 c.c. was used for the hydrolysis. The amount of starch used for the lots was 0·25 grms., and for lots with gelatinised starch this quantity was treated with a few c.c. of water and was subsequently heated on a sand-bath. The hydrolysis was allowed to proceed for 24 hours and at the end of this period the action was checked by the addition of a few drops

of strong alkali (sodium hydroxide) and subsequently boiling the mixtures on a sand-bath for a few minutes. During the period of reaction the fermentative changes were checked by the addition of a few drops of toluene to the reacting mixtures. After making the volume to 100 c.c. the mixtures were filtered through a dry filter paper and were subsequently titrated against standardised Benedict's solution. The activity of the enzymes has been expressed in terms of mgs. of dextrose formed from equal quantities of starch under the same conditions of temperature, dilution of the extract (strength of the extracts), etc. The following are the results obtained in these trials:—

TABLE XIX

Amount and extent of hydrolysis effected by the enzyme of the resting seeds and that of the germinated seeds, under similar conditions in terms of mgs. of dextrose formed from gelatinised and ungelatinised rice starch

Lot No.	Details of the lots	Dextrose formed	Duplicate
		Mgs.	Mgs.
I	Blank with extract of resting seeds . . .	39·24	39·24
II	Blank with extract of germinated seeds . .	44·44	44·44
III	Blank with extract of 0·5 per cent diastase absolute	0·0	0·0
IV	Ungelatinised starch *plus* extract of resting seeds .	64·51	64·51
V	Ungelatinised starch *plus* extract of germinated seeds.	68·68	69·44
VI	Ungelatinised starch *plus* extract of 0·5 per cent diastase absolute.	Traces	Traces
VII	Gelatinised starch *plus* extract of resting seeds .	102·5	105·26
VIII	Gelatinised starch *plus* extract of germinated seeds	312·5	307·6
IX	Gelatinised starch *plus* extract of 0·5 per cent diastase absolute.	222·2	222·2

From the above table we find that the extracts of both the resting and germinating seeds show the same amount of dextrose and that the extract of germinated seeds shows a little more of the same than the extract of resting seeds. Thus for the former we have 44·44 mgs. and for the latter we have 39·24 mgs.

of dextrose. This means that the resting seeds do contain some amount of dextrose. In the case of 0·5 per cent extract of diastase absolute, a blank experiment with the same gave no test for reducing sugars. Going now to the fourth lot we find that 64·5 mgs. of dextrose have been formed from 0·5 grms. of rice starch by the action of 20 c.c. of extract of resting seeds for a period of 24 hours. This proves the existence of an enzyme in the resting seeds, which is capable of attacking ungelatinised starch to some extent. By comparing the figure obtained for the first lot *i.e.* (for the blank with extract of resting seeds—39·24 mgs.) with that of the fourth one we find that the latter gives us an increase of nearly 25 mgs. which is clearly due to the action of the enzyme of the resting seed on ungelatinised starch. Going now to the fifth lot we find that the figure for the dextrose here is slightly higher than that for the previous one, the increase being a little more than four mgs. This increase is. therefore, due to the slightly greater activity of the enzyme of the germinated seeds and this activity may be probably due to the better conditions of temperature and moisture that are given to the germinating seeds.

It had already been pointed out in the Introductory part of this section that Brown and Morris [1895] are of opinion that the resting seeds do not contain any "starch-transforming body" and that "it is entirely a product of germination". However in the light of the results obtained in the present work it can be safely said that though slightly less active than the enzyme of the germinating rice seeds the resting seeds do contain such a starch-transforming body and that during the process of germination this body increases in its activity. These results are supported by the work of Kjeldahl [1880] who has actually proved the existence of "An active amylo-hydrolytic enzyme in ungerminated barley." Brown and Morris are of opinion that though this enzyme as shown to be present in ungerminated barley has a considerable saccharifying power yet it has "scarcely any influence in liquefying starch paste." Though in the present paper the actual liquefying power has not been studied yet it can be safely said from the results obtained that the enzyme of the resting seeds does act on ungelatinised rice starch and it could not be so if it had no liquefying power. Or it may be that even this enzyme may be a complex of two or more co-enzymes one of which may be with some power in liquefying the starch to some extent which is then attacked by the other and turned into dextrose.

Going now to lot six we find that we hardly get any hydrolysis and this means that an extract of diastase absolute (0·5 per cent) is not able to act on ungelatinised starch as the other two do, and slight traces of dextrose are formed in this case. This is supported by Brasse [1885] who says that "Commercial diastases have no effect on crude or uncooked starch, and this is due to the alteration that they undergo in the process of extraction."

Lot seven which represents the action of the extract of resting seeds on gelatinised starch, shows 102·5 mgs. of dextrose, and 105·26 mgs. as a duplicate of the same. Lot eight, which stands for the action of the extract of germinated seeds on gelatinised starch shows 312·5 mgs. of dextrose and 307·6 mgs. as its duplicate. Lot nine gives 202·2 mgs. and this stands for the action of diastase absolute (0·5 per cent) on gelatinised starch. This means that an extract of

germinating seeds is more active than that of the resting seeds. Comparing the figures of lots seven and eight with those of nine we find that an extract of diastase of 0·5 per cent strength has greater action on gelatinised starch than an extract of resting seeds (strength 30 per cent) though an extract of germinated seeds has greater action still than the above two. Similarly an extract of germinating seeds gives 210 mgs. and about 110 mgs. more of dextrose than an extract of resting seeds and an extract of diastase (0·5 per cent strength) respectively when they are allowed to act on equal quantities of rice starch. The following are some of the conclusions drawn from the results obtained in the present work:—

(1) Both the germinated and the resting seeds contain some amount of dextrose and it appears that it is formed from the action of the enzyme present in the seed. The amount present in the germinated seeds is slightly greater and this is probably due to the increased activity of the enzyme during germination.

(2) The enzyme of the resting seeds does act on ungelatinised starch, though of course the action is decidedly less when compared to that of the enzyme present in the germinating seeds. Diastase absolute however is not able to act on ungelatinised starch.

(3) The action of all the three enzymes is greater on gelatinised starch than on an equal amount of ungelatinised starch.

(4) The enzyme of the germinating seeds appears to have the greatest action on gelatinised starch; then comes the diastase and last is the enzyme of the resting seeds.

(5) The action of the extract of resting seeds on ungelatinised starch proves that it has some power of liquefying the starch, or it may be that this enzyme is a complex of two or more "co-enzymes" one of which liquefies the starch and the other turns it into dextrose.

After proving the existence of an amylo-hydrolytic enzyme in the resting seeds it is easy to understand the difference between new and old rice grains. We have already seen that this enzyme has the power of attacking both gelatinised and ungelatinised starch of the rice grain and it, therefore, appears that if the enzyme remains active during the resting period of the grains it may, to a certain extent, attack the starch that is present in it. We have seen that the starch and also the flour of the old rices undergo greater action when treated with different hydrolysing agents, than the starch and flour of new ones. This easy susceptibility of the old rices to the various treatments appears to be not merely due to a physical disintegration of the grain into simpler products, during the period of storage, but also due to a certain enzymic action during this period. We have seen that during the resting period the rice and its starch change some of their physical properties, and they are attacked to a greater extent by alkalies when treated with them. It is also noticed that old rices and their starches undergo greater disintegration when boiled with water than the new rices and the starches prepared from them.

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THE MECHANICAL ANALYSIS OF LATERITIC SOILS

2. A NOTE ON THE APPLICABILITY OF THE HYPOBROMITE METHOD

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Troell [1931] has developed a new method for the mechanical analysis of soils using sodium hypobromite. The method appears to be a promising general method and has an advantage over the popular International Soda method in that the agent (sodium hypobromite) for oxidation of organic matter in the new method, unlike that (hydrogen peroxide) in the International Soda method, is unaffected by the temperature of tropical countries.

In order to see if the hypobromite method was suitable for the mechanical analysis of Indian lateritic soils, a few such soils were analysed by this method and the results compared with those obtained by the International Soda method.

TABLE I

Percentage of clay obtained by different methods

Soil No.	3	4	5
Sodium hypobromite method	15·3	14·0	11·4
International Soda method	19·2	16·3	16·6

It is seen from above that in the case of soil Nos. 3, 4, and 5 the hypobromite method gave low clay figures, in other words, the dispersion in the case of these soils was incomplete.

In the first paper of this series [Chakraborty and Sen, 1932] it has been shown that these lateritic soils refused to disperse fully unless they were shaken for 24 hours in a suspension of pH about 10·5. In the hypobromite method shaking for half an hour is recommended with a suspension whose reaction may be slightly alkaline. To test, therefore, whether dispersion improved in the case of the hypobromite method, determinations were made employing 24 hours'

shaking and making the suspension more alkaline. The results are given in Table II below:—

TABLE II

Percentage of clay obtained by different dispersion treatments

Soil No.	International Soda method	Hypobromite method ; ½ hour shaking	Hypobromite method ; 24 hours' shaking	Hypobromite method ; 24 hours' shaking pH 10·5
3	19·2	15·3	16·7	19·1
4	16·3	14·0	15·0	17·3
5	16·6	11·4	12·9	15·2
6A	..	32·8	39·8	45·7
19	..	17·0	24·0	28·9

Comparison of results given in column 4 of the above table shows that better dispersion of soils was obtained by including the standard shaking procedure extending over 24 hours in the hypobromite method. Even then the amount of clay so obtained is smaller than that given by the International Soda method. But when the suspensions were made more alkaline and shaken for 24 hours the amount of clay thus obtained is practically equal to that given by the International Soda method.

It is clear, therefore, that the dispersion of some of the Indian lateritic soils by the hypobromite method is not complete unless the following modifications are introduced:—

(1) Addition of sufficient quantity of alkali so that pH of the final suspension is 10·5.

(2) A larger period of shaking than recommended in the original method.

But the greatest objection to the hypobromite method is that it requires the use of large quantities of obnoxious and injurious chemicals like bromine and ammonia and therefore quite unfit for inclusion in routine practice.

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3. A NEW METHOD USING ALKALINE PERMANGANATE FOR OXIDATION OF ORGANIC MATTER

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A method termed the "Direct Soda Method" for mechanical analysis of Indian lateritic soils was developed and published by the author and Sen [1932]. The essential feature of this method consists in dispersing the soil directly by shaking with water made alkaline, pH about 10·5, with sufficient caustic soda. This method was evolved in the light of the fact that the majority of Indian lateritic soils contained little organic matter, calcium carbonate and exchangeable bases so that the pre-treatment of these soils with hydrogen peroxide and hydrochloric acid as recommended in the International method was not necessary but it was shown that the dispersion medium should have a pH about 10·5. In exceptional cases where lateritic soils contained more than 2 per cent of organic matter, it was recommended to include in the method the pre-treatment with hydrogen peroxide so as to remove humus. It may be of interest to mention here that almost simultaneously with the publication of our paper [Chakraborty and Sen, 1932] embodying these results the Imperial Bureau of Soil Science brought out their Technical Communication No. 26 [Robinson, 1933] in which it was stated that while destruction of humus was always advisable to ensure complete dispersion, results of mechanical analysis of a large number of soils including laterites indicated that where less than one per cent carbon was present the depression of the clay figure due to non-removal of the organic matter was not serious. This statement corroborated our conclusion since one per cent organic carbon might be considered as equivalent to 1·72 per cent organic matter [Read and Riddgell, 1922].

It was however felt that a method for mechanical analysis should be free from limitations and that attempt should be made to find one general method for dispersion of lateritic soils in which organic matter is present in large or small quantities. Furthermore the use of hydrogen peroxide which is costly and unstable in India should be avoided. Accordingly a search was made for a suitable agent for the oxidation of organic matter. Sodium hypobromite as recommended by Troell [1931] was tried [Chakraborty and Sen, 1935] but it was considered unsuitable for inclusion in routine practice. Eventually alkaline permanganate was found to be a satisfactory agent and a new method for the mechanical analysis of all classes of lateritic soils has been developed using this agent as reported below.

Use of alkaline permanganate in oxidation of organic compound is well known [Bigelow, 1919, 1922]. Liquid fatty acids are also oxidized by alkaline permanganate and hydroxylated acids are obtained [Lewkowitsch and Warburton,

1921]. Fallot [1924] made use of potassium permanganate as an oxidizing agent for estimation of humus in soil after extraction of the same with caustic potash. So at the first instance 4 soils of lateritic type containing organic matter from 2·4 to 10 per cent were heated with a solution of potassium permanganate in presence of caustic soda and it was found that in duplicate experiments with a particular soil almost equal quantities of alkaline permanganate were decolorised. The reaction involved in the process of oxidation may be represented thus:—

$$2\,KMnO_4 + KOH + H_2O = 2MnO_2 + 3\,KOH + 3\,O$$

The oxygen liberated oxidises the organic matter. The hydrated manganese dioxide produced as a result of reaction and incorporated with the soil is removed by adding gradually a concentrated solution of sodium bisulphite in the soil acidified with hydrochloric acid. The soil is filtered, washed with sodium acetate solution until free from manganese and sulphate, and finally alkalised to pH 10·5 with normal sodium hydroxide and shaken.

Soil samples

Samples of soils used in this investigation represent the typical Indian laterites. Silica sesquioxide ratios of these samples vary from 1 tc 2 and all of them contain very little calcium carbonate and exchangeable bases.

Solutions used are:—

(1) Normal sodium hydroxide.

(2) Normal potassium permanganate prepared by dissolving 31·6 grms. of permanganate in water and making up to a litre; 50 c. c. of this solution is capable of oxidising ·09 grm. of organic carbon in an alkaline medium.

(3) Normal hydrochloric acid.

(4) 25 per cent sodium bisulphite solution, 5 c. c. of the freshly prepared solution dissolves manganese dioxide produced from 50 c.c. of the above permanganate solution. This solution should not be preserved.

(5) Acidified sodium acetate solution prepared by dissolving 100 grms. of sodium acetate (crystalline) in a litre of water and just acidified before use with normal hydrochloric acid adding a few drops of methyl orange as internal indicator. Calcium sulphate is about 4 times more soluble in this reagent than in water. Hydrated manganese dioxide is also slightly soluble in it.

Experimental

(*a*) *Oxidation of organic matter*

Twenty grms. of air-dry soil are taken in a 600-c. c. Pyrex beaker to which 50 c. c. of permanganate solution and 5 c. c. normal sodium hydroxide are added

successively. The beaker is then placed over a sand-bath and the mixture stirred frequently. As soon as the permanganate colour disappears a fresh addition of 50 c. c. of the permanganate solution is made. This is repeated until the colour lasts for about half an hour after a fresh addition. In soils containing large quantities of organic matter 100 c. c. permanganate may be decolorised within 15 minutes. After the permanent colour has been obtained, 25 c. c. permanganate and 5 c. c. normal sodium hydroxide are added and the reaction completed by heating for half an hour more. Oxidation of humified organic matter is taken as complete when 25 c. c. of permanganate is not decolorised after heating for 30 to 40 minutes. It has been observed that soils containing more than 10 per cent organic matter require less than two hours and a half for this operation while in the case of the majority of the soils reported here oxidation was found to be finished within an hour and a half.

(*b*) *Removal of calcium carbonate*

Although Indian lateritic soils do not contain sufficient calcium carbonate and exchangeable calcium to interfere with direct dispersion with sodium hydroxide, it is advisable to include a process for removing calcium carbonate so that the present method may be applicable to all types of soil. After oxidation of organic matter is complete, the beaker is taken off the sand-bath and allowed to cool. Enough hydrochloric acid is then run in from a burette until evolution of carbon dioxide ceases and the mixture is rendered slightly acid to blue litmus paper. A further addition of 50 c. c. normal hydrochloric acid is made and water is added if necessary, to make the mixture about $N/5$ acid. The contents of the beaker are frequently stirred with a glass rod for about an hour and allowed to stand until the soil settles. The supernatant liquid is passed through a filter and the residue is washed with about 50 c. c. water by decantation. Soils containing unusually large quantities of calcium carbonate should be given a further washing with 50 c.c. of water.

(*c*) *Separation from hydrated manganese dioxide*

Ten c. c. of normal hydrochloric acid for each 50 c. c. of permanganate used for oxidation and 20 c. c. in excess are run into the beaker containing the washed soil from (*b*) and sufficient water is added to adjust the strength of the acid to about $N/2{\cdot}5$ 3 to 4 c. c. lots of 25 per cent sodium bisulphite solution are then added but the contents of the beaker are shaken by hand after each addition. This process is continued until all manganese dioxide is dissolved when the soil looks free from any dark colour. The material in the beaker is allowed to settle and the supernatant liquid is passed through the same filter used in (*b*). Any black particle of manganese dioxide that might have adhered to the filter in course of filtration in (*b*) will be dissolved when it comes in contact with this decanted liquid. Otherwise a dilute solution of bisulphite is to be passed through the filter. To ensure complete removal of manganese dioxide 5 c. c. of normal hydrochloric acid solution and 1 to 2 c. c. of bisulphite solution may be added to the residue in the beaker and shaken for

a few minutes. Treatment with bisulphite in presence of acid should not be carried for a longer period than necessary for removal of manganese dioxide. This operation (*c*) does not require more than 30 to 40 minutes even in soils containing more than 10 per cent organic matter.

(d) Washing free from manganese and sulphate

The soil is transferred to the filter and washed with about 10 c. c. water and then with sodium acetate solution allowing the filtrate to drain off completely before a fresh addition of the acetate solution is made. Washing with this reagent is continued until the filtrate is free from sulphate and manganese. Manganese may be tested by boiling a few drops of the filtrate with a little bromine water and then again boiling with a few drops of ammonium hydroxide when a black precipitate indicates presence of manganese. The presence of sulphate may be detected by the usual barium chloride test in the hot. For most soils washing with 200 to 300 c. c. of acetate has proved quite sufficient. In exceptional cases 400 to 500 c. c. may be required. Finally the soil is washed twice with about 25 c. c. water through a fine jet so as to remove the adhering sodium acetate from the beaker and the filter paper as far as possible.

It was found that filtration and washing in Buchner-funnels under pressure is much more rapid and efficient than those carried out in ordinary funnels. For fixing up the filter paper with the Buchner funnel we have used paraffin in place of Durofix cement (cellulose acetate) and found it to be quite satisfactory.

(e) Dispersion and sampling

After the soil has been washed and drained, it is transferred into a 500-c. c. bottle and 8 c. c. of normal sodium hydroxide is added for every 20 grms. of soil used. The bottle is shaken in an end-over-end-shaker at the rate of about 40 revolutions per minute for six hours. The contents of the bottle are made up to 2 litres and subjected to pipette sampling for determination of clay and silt. Coarse sand and fine sand are estimated by sieving and sedimentation respectively. The soil need not be rubbed while passing through 0·2 mm. (No. 70 I. M. M.) sieve.

(f) Determination of loss on solution

An aliquot part generally 1/10 to 1/5 of the filtrate from *b* to *d* (rendered free from permanganate colour if necessary with a few drops of bisulphite) is boiled with a little concentrated nitric acid to oxidize any ferrous iron and sodium bisulphite present in the filtrate. A modified method of determination of sesquioxide in presence of a large quantity of manganese dioxide is followed. The essential feature of this method lies only in using a larger quantity of ammonium chloride and a limited quantity of ammonium hydroxide for precipitation of iron and aluminium. This method has been developed by Lundell and Knowles [1923] and described by Treadwell and Hall [1930] thus:—

"To the acid solution containing 5 grms. of ammonium chloride and not more than 0·2 grm. of iron and aluminium cations in 20 c. c. of solution, add a

few drops of a 0·2 per cent solution of methyl red in alcohol and heat just to boiling. Carefully add normal ammonium hydroxide solution drop by drop until the colour of the solution changes to a distinct yellow. Boil for 2 minutes and filter promptly. Wash with hot 2 per cent ammonium nitrate solution until free from chloride.....As good a separation of iron and aluminium from manganese is accomplished in this way as by a single basic acetate precipitation. It is advisable to dissolve the precipitate and repeat the precipitation when the precipitate on ignition is likely to weigh more than 0·1 grm. or when a 9 cm. filter is more than half full or the precipitate".

Experimental results and discussions

In view of the fact that International Soda method is so far considered as the most satisfactory and rapid method of mechanical analysis applicable to a large variety of soils it was decided to carry out comparison of the clay and clay + silt figures obtained by this method and the method under investigation using 18 typical Indian lateritic soils. It has been pointed out by Robinson [1933] and rightly so that in the case of gypseous soils the International Soda method also needs modification. As there is no gypsum present in the soils under examination, the International Soda method is expected to yield satisfactory results.

TABLE I

Percentage of clay and clay+silt in air-dry soil

Soil No.	Lab. No.	Locality	Loss on ignition	International Soda method H_2O_2-HCl-NaOH (One per cent suspension of 20 grms. soil shaken for 24 hrs. with 8 c. c. *N* NaOH)			Alkaline permanganate method (One per cent suspension of 20 grms. soil shaken for 6 hrs. with 8 c. c. *N* NaOH)		
				Clay	Clay + silt	Loss on solution	Clay	Clay + silt	Loss on solution
1	34	Madras	16·5	44·4	57·1	0·4	44·2	55·4	1·3
2A	58A	Bombay	17·5	39·4	53·6	0·43	37·7	53·8	0·97
3	64	Burma	10·8	18·4	23·3	1·6	19·4	24·7	1·2
4	74	Orissa	5·8	15·8	35·0	0·92	16·9	36·5	1·3
5	24	C. P.	9·7	11·8	24·1	0·14	12·1	25·4	0·4
6A	30A	Madras	25·2*	48·4	59·0	2·2	49·7	62·8	1·4
7	80	Assam	11·9	29·2	46·8	0·6	28·4	46·6	0·6
10	11	Bihar	5·5	22·8	36·8	0·3	23·0	37·7	0·5
14	26	Madras	3·8	11·7	14·7	0·2	11·7	14·8	0·3
15	20	Burma	4·0	12·1	18·5	0·35	12·2	18·7	0·47
16	38	Bengal	4·7	18·2	28·2	0·8	18·3	28·2	0·45
17	44	Bengal	2·8	9·7	24·3	0·24	9·8	24·4	0·4
19	88	Assam	22·7	31·3	61·9	3·7	33·7	65·7	1·7
20	56	Bombay	18·3	45·2	59·6	1·0	45·4	61·5	0·94
21	60	Burma	13·5	48·2	61·8	0·7	46·8	60·1	1·2
22	66	Bihar	3·5	14·0	22·7	0·5	14·3	22·9	0·6
23	76	Orissa	11·3	41·2	62·8	0·7	40·5	63·7	0·96
24	86	Assam	4·3	12·5	22·2	0·46	12·7	22·2	0·6

*Containing 10 per cent organic matter

A perusal of Table I clearly indicates that the agreement between the two sets of figures by the two methods are on the whole very good. The alkaline permanganate method is therefore as efficient as the International Soda method in effecting maximum dispersion of soil. Further, the percentage loss on solution in the permanganate method did not exceed that in the International Soda method.

Time of shaking required for complete dispersion

Six soils rich in organic matter were chosen and their clay figures were estimated by the alkaline permanganate method shaking the final suspension to varying lengths of time from 3 to 24 hours. The results are given in Table II below from which it will appear that it is not necessary to shake for more than six hours with this method.

Table II

Percentage of clay in air-dry soils

Soil No.	Hours of shaking			
	3	6	12	24
2A . . .	37·9	37·7	37·9	38·3
3	19·3	19·4	19·4	19·2
5	12·6	12·1	12·6	13·4
6A . . .	47·1	49·7	49·4	49·9
19	33·9	33·7	33·5	33·7
20	45·1	45·4	45·6	46·3

Soils to which large quantities of calcium carbonate and calcium sulphate have been added

As no soil rich in either lime or gypsum was available in this laboratory it was thought to test the applicability of alkaline permanganate method in samples of soils mixed with large quantities of calcium carbonate and calcium sulphate. It is admitted that the behaviour of these soils may not be the same as that of soils rich in lime or gypsum in the natural state, but it is thought that some idea as to the interference to dispersion, if any, due to lime and gypsum may be obtained from experiment with these mixed soils. 20-grm. lots of four soils were taken. One lot was left untreated while 4 grms. of calcium carbonate were added to each sample of the second lot and 4 grms. of calcium carbonate + 2 grms. of calcium sulphate to each sample of the third lot.

The results of analysis of these soils by the alkaline permanganate method are given below.

TABLE III

Soil No.	Loss on ignition	Per cent clay (Untreated soil)	Per cent clay soil + calcium carbonate	Per cent clay soil + calcium carbonate + calcium sulphate
2A . . .	17·5	37·7	37·7	36·7
3	10·8	19·4	19·0	19·1
6A . . .	23·2	49·7	48·4	48·6
19	22·7	33·7	32·9	34·0

It is clear from the table that maximum dispersion is obtainable by the alkaline permanganate method whether the soil contains calcium carbonate and calcium sulphate or not. This is probably due to the fact that greater part of the gypsum present in the soil is transformed into calcium carbonate as a result of reaction between calcium sulphate and alkali carbonate produced during oxidation of organic matter with alkali permanganate. The reaction may be represented thus:—

$$Na_2CO^3 + CaSO_4 = Na_2SO_4 + CaCO_3$$

The calcium carbonate formed along with that originally present in the soil is removed by the subsequent hydrochloric acid treatment followed by washing. On the other hand, washing with sodium acetate solution included in the method also helps to remove gypsum to a great extent, for the solubility of calcium sulphate in the acetate solution is found to be 4 times greater than in water. It must be pointed out however, that for soils containing gypsum sodium hydroxide as demanded by the above equation (15 c. c. normal caustic soda for each one grm. calcium sulphate) should be added gradually in addition to the quantity of alkali used with permanganate to remove organic matter. Coarse gypsum present in gypseous soils should be removed by passing the soil through a 70-mesh sieve after oxidation with alkali permangnate and before addition of hydrochloric acid [Robinson, 1933].

Finally it may be mentioned that although the alkaline permanganate method has been shown here to be quite suitable for the mechanical analysis of Indian lateritic soils, it is expected that the method will be found equally suitable for all other types of soil.

SHORT DESCRIPTION OF THE ALKALINE PERMANGANATE METHOD FOR THE MECHANICAL ANALYSIS OF LATERITIC SOILS

Twenty grms. of soil are taken in a 600-c. c. beaker to which 50 c. c. of 3·16 per cent potassium permanganate and 5 c. c. of normal caustic soda are

added. The beaker is then placed on a sand-bath and the mixture stirred frequently. As soon as the colour disappears a fresh 50 c. c. of permanganate solution is added. This is repeated until the colour lasts for half an hour when a further addition of 25 c. c. permanganate and 5 c. c. normal caustic soda is made and the whole thing heated again for 30 to 40 minutes to bring oxidation to a finish.

The beaker is then allowed to cool. Sufficient normal hydrochloric acid is run until evolution of carbon dioxide ceases and the mixture becomes slightly acid. Further 50 c. c. of normal hydrochloric acid is added and the strength of the acid in the beaker is adjusted to $N/5$ with water. The contents of the beaker are frequently stirred for one hour and allowed to settle. The supernatant liquid is filtered off and the residue in the beaker is washed with about 50 c. c. water by decantation to remove greater part of calcium salts.

Ten c. c. of normal hydrochloric acid for each 50 c. c. of permanganate used and 20 c. c. in excess are poured into the beaker and an equal quantity of water is added. Twenty-five per cent solution of sodium bisulphite added to the material in the beaker in doses of 3 to 4 c. c. stirring frequently after each addition until all manganese dioxide is dissolved.

The mixture is allowed to settle and the supernatant liquid is poured on the filter used previously. The residue in the beaker is transferred to the filter and washed with a little water and then with 10 per cent sodium acetate solution, just acidified with hydrochloric acid, until the filtrate is free from manganese and sulphate. The soil is drained and finally washed twice using 25 c. c. water each time.

The residue in the funnel are transferred to a 500-c. c. bottle by means of a jet of water and shaken for 6 hours in a rotary shaker moving at a speed of about 40 revolutions per minute with 8 c. c. of normal caustic soda.

The soil suspension is made up to 2 litres and subjected to pipette sampling.

Summary

(1) A new method for the mechanical analysis of lateritic soils has been developed using alkaline permanganate to remove organic matter.

(2) The results for clay and clay plus silt obtained by this method in the case of 18 lateritic soils from all over India were comparable to those obtained by International Soda method.

(3) The loss on solution in this method is not greater than that in the International Soda method.

(4) Shaking for six hours in a rotary shaker is sufficient for complete dispersion of soil by this method.

(5) This method without modification was able to disperse completely four lateritic soils in which large quantities of calcium carbonate and sulphate have been added.

(6) The method is described fully.

(7) Advantages of the method are:—

(i) Alkaline permanganate is equally suitable in temperate and tropical countries where hydrogen peroxide is less stable.

(ii) It can be used in manganiferous soils where hydrogen peroxide is required in large quantities or is useless.

(iii) Time required for oxidation does not exceed three hours even in soils containing large quantities of organic matter (*e.g.*, 10 per cent). On the other hand hydrogen peroxide and sodium hypobromite require much longer time for the purpose.

(iv) Obnoxious and injurious chemicals like bromine, as in the method using sodium hypobromite, are avoided.

(v) This reagent is much cheaper than either hydrogen peroxide or sodium hypobromite.

(8) Although the alkaline permanganate method has been shown here to be quite suitable for the mechanical analysis of Indian lateritic soil, it is expected that the method will prove equally suitable for all other types of soil.

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STINKING SMUT (BUNT) OF WHEAT WITH SPECIAL REFERENCE TO *TILLETIA INDICA* MITRA

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(With Plates I–VII and two text-figures)

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I. Introduction

Stinking smut or bunt of wheat was until recently believed to be caused by two closely related species of Tilletia *viz.*, *T. caries* (DC.) Tul. [*T. tritici* (Bjerk.) Wint.] and *T. foetens* (Berk. and Curt.) Trel. (*T. laevis* Kuehn.), but diseased seeds of certain wheat varieties collected at Karnal (Punjab) in May 1930 showed the presence of an undescribed species of this genus differing from *T. caries* and *T. foetens* in several respects. It was named by the author as *T. indica* and a brief description has already been published [Mitra, 1931, 1].

In India, three species of Tilletia are known to occur and their distribution is as follows:—

T. caries.—Gilgit (Kashmir), Sopor (Kashmir), Simla Hills and Kangra (Himalayan range).

T. foetens.—Gilgit (Kashmir), Sopor (Kashmir), Jubbal (Simla Hills) and Baluchistan (Himalayan and Sulaiman Mountain ranges).

T. indica.—Karnal, Lahore, Amballa, Jaranwalla, Lyallpur and Ropar (Punjab) and Peshawar (North Western Frontier Province).

It will be noticed from the distribution of the three species that so far as it is known at present, bunt caused by *T. caries* and *T. foetens* is confined to the cooler regions of the Himalayas and Baluchistan, while *T. indica* occurs only in the plains of the Punjab and the North Western Frontier Province.

Howard and Howard [1909] recorded the occurrence of bunt on wheat at Lyallpur. *T. indica* has been found at that locality and the neighbouring districts in the plains of the Punjab during the last four years. It is likely that the bunt observed by Howard and Howard as early as 1908 was caused by this species.

The author has not been able to find any specimen of *T. indica* in the herbarium collection of bunts of wheat preserved at the Imperial Institute of Agricultural Research, Pusa. This shows that this bunt was never even collected before.

It has been found that low temperatures are most favourable for the infection of susceptible wheats by *T. foetens* and *T. caries*. At temperatures much above 10°C., the percentage of infection falls off rather rapidly and few seedlings grown at temperatures higher than 20°C. are infected. Munerati [1922] obtained 90-100 per cent of bunt with *T. caries* and *T. foetens* in wheat germinated at 7-8°C. while the same varieties gave 1·4-12 per cent infection when germinated at 18-20°C. The same author [Munerati, 1923] found that seeds germinated at 10-12°C. gave 13·2-39·3 per cent infection while the same two varieties produced 0-1·4 per cent bunt at 22-25°C. Similar results were obtained by Volkart [1906], Heuser [1922] and Hungerford [1922]. Faris [1924] states that infection does not take place at 20°C., 25°C. and 30°C. with *T. caries* and very rarely at 20°C. and 25°C. with *T. foetens*. He obtained successful infection with *T. caries* at temperatures between 5 and 15°C. (mostly at 5-10°C.) and with *T. foetens* between 5 and 25°C. (mostly at 10-15°C.) and very rarely at 25°C. Tisdale, Leighty and Boerner [1927] in their studies of the distribution of *T. caries* and *T. foetens* have also shown that temperatures ranging from 5 to 15°C. are very favourable for infection.

The conclusion drawn by these authors is that *T. caries* is more generally distributed throughout the cooler regions and that *T. foetens* can tolerate slightly higher temperatures. Thus it is clear that for infection with *T. caries* and *T. foetens* the temperature should not be above 15°C. and that temperature plays an important part in the distribution of these bunts. *T. caries* and *T. foetens* require a much cooler climate to thrive in than *T. indica*. That seems to be the reason why *T. caries* and *T. foetens* are not found on the plains of North-West India where this low temperature generally does not occur at the time of sowing or harvesting wheat and this accounts for their absence from localities where the average temperature is above 20°C. at the time of sowing and above 30°C. at the time of harvesting. The occurrence of *T. indica* on the plains indicates that this species can stand warm weather. At Karnal the average maximum temperature at the time of sowing of wheat was 34·5°C. during 1932-33 and 29·5°C. during 1933-34. Though the minimum goes down to 14°C., the number of hours per day having more than 20°C. temperature is far more than in the hills.

Further, it may be mentioned that the weather following the harvest in the hills where *T. caries* and *T. foetens* occur is cool whereas on the plains of North-West India where *T. indica* alone occurs, the wheat season is followed by a hot summer.

II. Losses due to stinking smut (bunt)

There is very little information available regarding the damage done to wheat by bunt in India. It appears however that *T. caries* and *T. foetens* do a good deal of damage to wheat in Kashmir, Baluchistan and in the Simla Hills. In 1933 it was reported from Jubbal (Simla Hills) that the damage due to *T. foetens* was from 30-40 per cent. In 1923 bunt was reported to be very bad at Peshawar and for the first time did considerable damage to both the bearded and beardless varieties. As no specimens were sent to Pusa, it was not possible to determine the species of Tilletia responsible for the loss. During 1932, it was again reported to have done considerable damage to the wheat crop, especially the beardless varieties, at Peshawar, and the loss was estimated to be up to 33 per cent in several cases. Specimens of bunted grain showed the presence of *T. indica* and most probably the species responsible for the loss during 1923 at Peshawar was the same, as the other two species have so far not been recorded from that locality.

T. indica was first observed at Karnal on certain hybrids (cross between Federation and Pusa 4 and Pusa 52), *viz.*, Pusa 4 × Fed. (1—1), Pusa 4 × Fed. (7—1), Pusa 4 × Fed. (97—1), Pusa 4 × Fed. (64—1). Pusa 4 × Fed. (3—1) = P. 165, Pusa 52 × Fed. (20—1) = P. 120, Pusa 52 × Fed. (38—1), Pusa 52 × Fed. (64—1). During 1931 it was further observed on Pusa 4, 52, 114, 115, 116, 117, Federation and Punjab 8A. During 1933, it appeared in a virulent form at Karnal on several of the wheat varieties Pusa 4, 12, 52, 114, 165 were slightly attacked, Pusa 80—5, 111. 120 had infection up to 2 per cent and Pusa 112 and 113 showed nearly 10 per cent Seed of Pusa 72—1—1 was threshed at the time of examination and showed more than 20 per cent diseased grain.

The estimate of the percentage of infection is based on the number of ears having bunted grain. The loss in yield thus is much less, as only a very few spikelets in an ear are attacked.

The presence of *T. indica* on Punjab 8A indicated the probability of its being present in other parts of the Punjab. Consequently an opportunity was taken to examine wheat in the Lahore grain market where it is stored in large quantities from different parts of the province and bunted grain was found to be fairly common. The disease is therefore fairly widespread throughout the plains of that province.

III. Symptoms produced by *T. indica*

The previous description of this fungus [Mitra, 1931] was based on a small collection of bunted grain picked out after threshing. The disease being rather severe during 1933 at Karnal, an opportunity was taken to make more detailed observations.

In a stool all the ears are not infected and in an ear all the grains are not bunted. Thus the same plant produces sound and bunted ears and the same ear, sound and bunted grain. A completely infected ear has never been observed, the sori being only produced in a few individual spikelets; generally 1—5 spikelets show bunted grain in an ear while the rest produce healthy grain. It is difficult frequently to detect diseased grain in an ear without breaking the spikelets, as the grain is hidden by the enclosing unaffected glumes. This is especially the case in unripe ears. When the grain ripens the diseased spikelets are more open, the outer glumes spread out giving more space to the inner glume and the palae to expand with the result that the bunted grain becomes visible between the glumes. In early stages, a careful examination of the embryo portion of the seed helps to detect the disease where a black spot on the grain is noticeable at the tip of the spikelet. Often the grain is very slightly infected at the tip and the presence of the fungus can be noticed only with the help of a lens. In badly infected spikelets the glumes spread apart and later on fall off, thus exposing the bunted grain which also falls to the ground with a slight disturbance and infects the soil. Plate I illustrates the presence of bunted grain in ears.

No smell was detected in the small samples previously collected and recorded [Mitra, 1931]. Because of the abundance of material collected during 1933 it was possible to find out that this bunt like the other two has also a distinct stinking smell resembling that of rotten fish and the smell is more pronounced when the bunted grain is crushed.

The bunt affects the kernels only partially, which are not swollen. The embryo tissue except in very severe cases is not destroyed though in a large number of cases only the embryo portion is infected. Generally the infection spreads to the tissue along the groove but the endosperm material lying along the smooth side of the grain is uninfected. Bunted grain has been found to germinate freely, the percentage of germination being as high as eighty-nine in several cases. Further, they germinate more quickly than the healthy grain. *T. indica* is more highly differential than *T. caries* and *T. foetens* which leave only the glumes and the epidermal tissues of the kernels uninfected. The various types of infection of the grain from mild to severe are illustrated in Plate II.

Sori are formed under the pericarp and testa above the aleurone layer of the endosperm and are brown to dark brown in colour (Plates III, IV and V figs. 1 and 2). The attack starts at the embryo end and runs along the groove leaving the endosperm intact covered by the whole or partly ruptured pericarp. Gradually the endosperm region is replaced by the fungus. At first the spores in sori are enclosed by the pericarp and cannot be seen from outside and only microscopic examination (transverse and longitudinal sections) reveals its presence (Plates IV and V, fig. 2). Later on the pericarp ruptures and the spore mass is exposed. (Plates III and V, fig. 1). A collection of bunted grain shows all stages of infection from very slight to severe.

IV. Morphology of *T. indica*

The spores bearing hyphae in a sorus give off pear-shaped bud clusters (Plate III and Plate V, fig. 1). These outgrowths increase in diameter and become rounded so that they are ultimately attached to the hyphae by a thin stalk. The membrane is gelatinous in young spores and disappears on maturity. The stalk or the thread of detachment sometimes remains attached to the spore. In transverse and longitudinal sections of diseased grains the fungal filaments and numerous spores are interspersed between the outer skin and the aleurone layer on the under-surface. The embryo remains quite intact though partially surrounded by numerous spores.

Spores when mature are dark brown, black in mass, spherical, subspherical or oval, 22—44 × 25—55 μ in diameter, average 32·5—40 μ. The proliferations of the thick epispore are reticulate, the ridges or scales are somewhat roundish, or bluntly projecting showing at the circumference of the spore a band 2 – 6 μ in width enveloped by a clear membrane probably gelatinous. Mixed with spores are numerous yellowish or sub-hyaline sterile cells, rounded, angular or pear-shaped, smaller in size than the spore, relatively thin-walled, and occasionally small brown cells occur which are smooth-walled and much smaller than spores. These in part represent undeveloped spores. The spores often have a papilla or thread of detachment. The spore mass dries up into pustules which are capable of being blown about by wind. Plate V, fig. 3, Plate VI, fig. 1 and Plate VII, figs. *a*, *b* and *c* illustrate the shape and details of spores.

The spores of *T. indica* differ considerably from *T. foetens* which has smooth-walled spores, much smaller in diameter and lighter in colour. Spores of *T. caries* resemble those of *T. indica* in the structure of their walls but they are much smaller in size and lighter in colour than those of the latter.

Spores of *T. caries* are dusty olive brown or rust coloured in mass while those of *T. indica* are black. In *T. caries* sterile cells are few while in *T. indica* they are numerous. In *T. caries*, the whole tissue is infected and except the outer coat everything is destroyed and partly infected grain is rare [McAlpine, 1910 and Heald, 1916] while in *T. indica* partly infected grain is very common and the whole tissue is seldom destroyed and completely infected grain is very uncommon.

V. Variation in spore size of *T. caries* and *T. indica*

Brentzel [1933] recently presented evidence to show that there existed a wide variation in the size and shape of spores and in the degree of spore-wall reticulations of *T. caries*. In the photograph which he published there were also present some unusually dark spores which appeared to resemble in appearance the spores of *T. indica*. It was thought advisable therefore to compare the spores of *T. indica* with those illustrated by Brentzel. Dr. Brentzel very kindly supplied the material from two collections, one from Ceres wheat (originally from Emmer) and the other from Kota wheat (originally

from Mindum, durum). For the sake of convenience the strain on Ceres is hereafter referred to as the Emmer strain and the other on Kota as the Durum strain.

For biometrical analysis of the spore sizes and their comparisons two hundred spores from each collection were measured. In addition to the two strains of *T. caries* supplied by Brentzel, a collection of *T. caries* from Gilgit (Kashmir) and two collections of *T. indica*, one from Karnal and the other from Peshawar, were included in the measurements. The frequency distributions for length and width (Figs. 1 and 2) and the biometrical constants are recorded in Tables I and II.

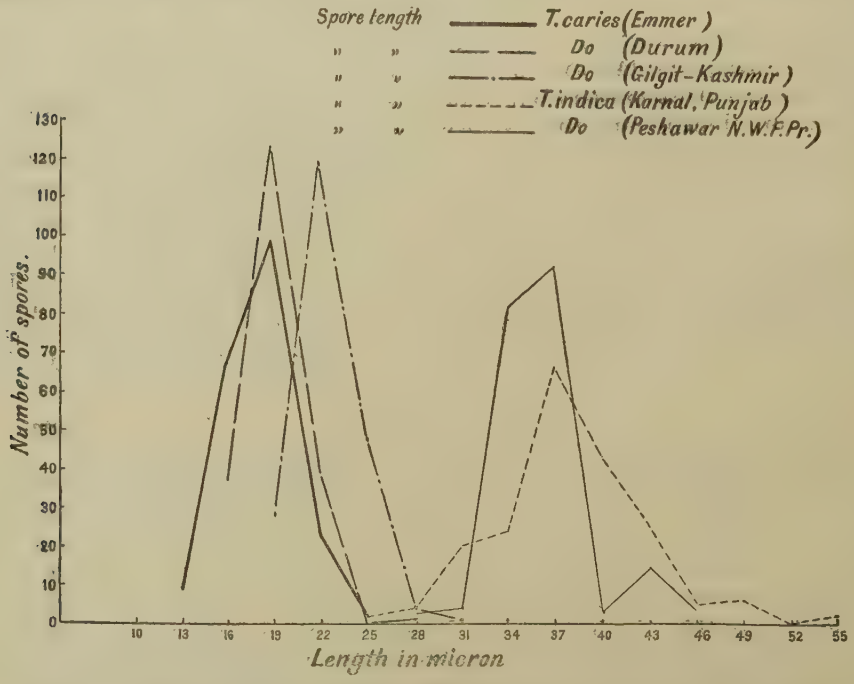

Fig. 1.—Comparison of spore length of three forms of *T. caries* and two forms of *T. indica*.

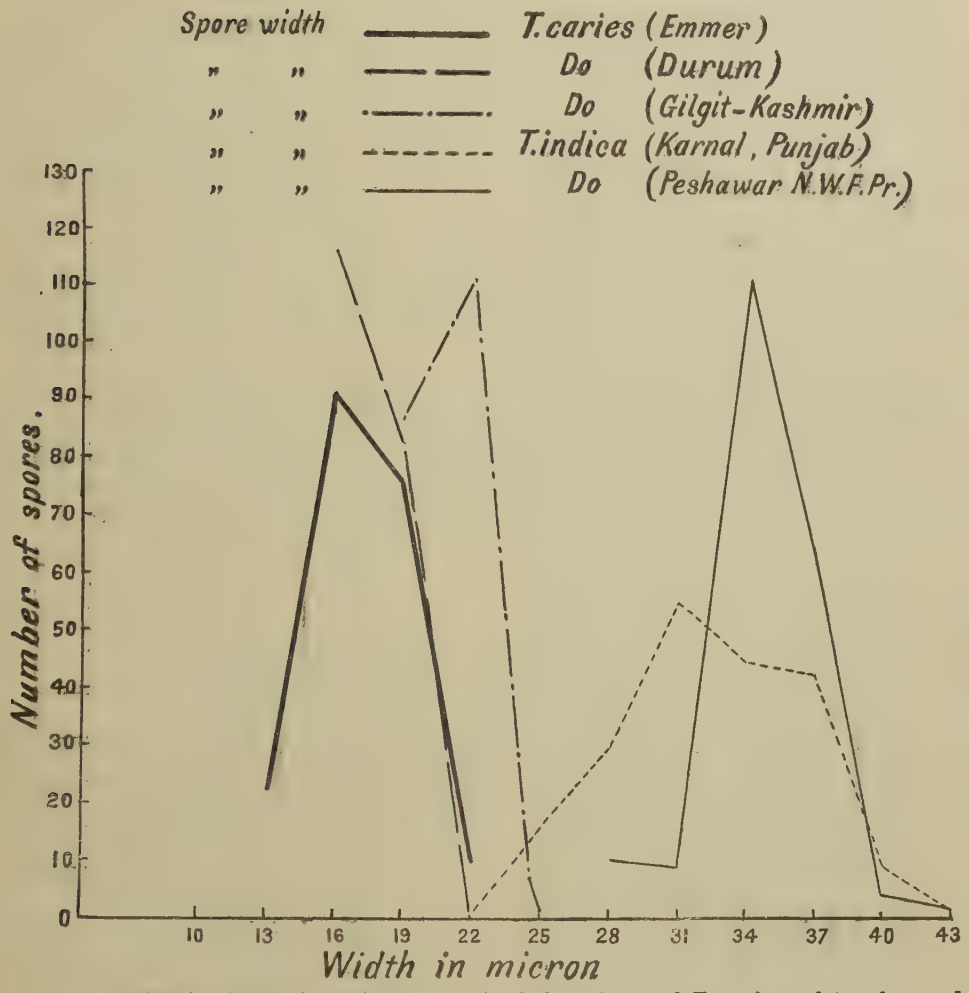

Fig. 2.—Comparison of spore width of three forms of *T. caries* and two forms of *T. indica.*

TABLE I

Variations and constants of spore length and width of three forms of T. caries

Bunt form	Spore length and width classes (in microns)							Range	Mode	Constants		
	13	16	19	22	25	28	31			Mean	S. D.	C. V.
Emmer L	9	67	99	23	2	...	...	12—25	19	18·1±·10	2·12±·07	11·7 ±·39
„ W	22	91	77	10	...	...	...	12—23	16	17·1±·09	2·05±·06	12·02±·40
Durum L	...	37	123	39	...	1	...	15·4—27·5	19	19·1±·08	1·75±·05	9·17±·30
„ W	...	116	83	1	...	...	...	15·4—22	16	17·3±·06	1·43±·04	8·28±·27
Gilgit L	...	...	28	119	48	4	1	18—30	22	22·6±·09	1·89±·06	8·42±·28
„ W	...	...	88	111	1	...	...	18—24	22	20·7±·05	1·25±·04	6·04±2·03

TABLE II

Variations and constants of spore length and width of two forms of T. indica.

Bunt form	Spore length and width classes (in microns)												Range	Mode	Constants		
	22	25	28	31	34	37	40	43	46	49	52	55			Mean	S. D.	C. V.
Karnal L	...	2	4	20	24	66	43	28	5	6	...	2	22—55	37	37·9±·22	4·75±·16	12·51±·42
„ W	1	16	30	55	45	43	9	1	...	...	...	...	22—42	31	32·5±·19	3·98±·13	12·28±·41
Peshawar L	...	...	3	4	81	91	3	14	4	...	...	...	27·5—46·2	37	36·2±·13	2·90±·09	8·02±·27
„ W	...	...	10	9	112	64	4	1	...	...	...	...	27·5—44	34	34·7±·10	2·24±·07	6·45±2·17

(L = Length, W = width)

From the data recorded in Tables I and II, it will be noted that the range of spore length and width of the Emmer strain is 12–25 μ ×12–23 μ with a mean of 18·3 μ ×17·1 μ, that of Durum strain is 15–28 μ ×15–22 μ with a mean of 19·1 μ ×17·3 μ and that of the Gilgit strain is 18–30 μ ×18–24 μ with a mean of 22·5 μ ×20·7 μ. The range in the case of the Karnal collection of *T. indica* is 25–55 μ ×22–42 μ with a mean of 37·9 μ × 32·5 μ and that of the Peshawar collection 27—46 μ × 27—44 μ with a mean of 36·2 μ ×347μ.

The above figures indicate therefore that apart from the differences already stated in the section on morphology, the spore sizes of the two species are absolutely different, that is, spores of *T. indica* are almost double the size of those of *T. caries*.

Furthermore, the data recorded in Tables I and II indicate that even within the species themselves, there is evidence of the occurrence of different physiologic forms. The mean length and width of spores from three collections of *T. caries* and two of *T. indica* showed considerable differences and in order to know whether the respective mean differences were statistically significant or not, Pearson's method of comparing the frequencies was applied. The results are recorded in Table III.

TABLE III

Probability for or against the different collections of T. caries *and* T. indica *being the same physiologic forms*

Fungus	Collections compared	Length or width	χ^2	Odds	Remarks
T. caries .	Emmer and Gilgit .	Length .	227·88	Odds against very high	Not same.
	,, ,, .	Width .	199·92	,,	,,
	Emmer and Durum.	Length .	27·32	,,	,,
	,, ,, .	Width .	32·60	,,	,,
	Durum and Gilgit .	Length .	188·04	,,	,,
	,, ,, .	Width .	229·12	,,	,,
T. indica .	Karnal and Peshawar	Length .	95·27	,,	,,
	,, ,, .	Width .	85·64	,,	,,

Odds in each and every case are very highly against their being the same physiologic forms. The Emmer, Durum and Gilgit strains of *T. caries* could only be considered therefore as physiologic forms. According to Stakman [1929] there are four general methods for recognising physiologic forms, cultural characters, physico-chemical reactions, pathogenicity on certain selected plants and sometimes morphology; but the spore size which is a characteristic and definite morphologic feature shows wide variations even within the same species in the above-mentioned cases. So far as the Peshawar and Karnal strains of *T. indica* are concerned, although physiological, physico-chemical and parasitic specialization studies have not so far been carried out, yet there are hardly any morphological differences except significant variation in their spore sizes to distinguish them as two different species. The writer however would like to adopt the more conservative view of this question and consider them as merely physiologic forms even though the spore size is so considerably different. Furthermore, the writer would like to emphasize that whereas spore length and width are definite characters of specific value, the differences should not be interpreted too narrowly. The writer has shown [Mitra, 1931,2] that saltants arise from monoconidial cultures of Helminthosporium sp., and that they show considerable variation in spore colour, septation, size, etc. Biometric analysis of spore measurements should therefore be taken as an additional criterion by which physiologic forms may be determined.

VI. Spore germination of *T. indica*

The most favourable conditions for profuse spore germination of *T. indica* have so far not been discovered. Freshly collected spores generally do not germinate and it appears they require a period of rest. Very few spores germinated under various conditions tried, such as, in liquid media like distilled, tap and rain water; one per cent solutions of cane sugar, asparagin, malt extract or soil extract. Even on solid media, such as, plain agar, Brown's synthetic agar, synthetic starch agar and potato juice agar, the germination was extremely poor.

Bunt was kept in ice from 10–45 days and tested weekly but did not show better germination; on the other hand material kept at 35°C. in an incubator for ten days germinated slightly better and more quickly than the frozen material.

Whenever the spores germinated, the germination took place at a temperature ranging from 15—25°C. Those kept at 10°C. or below or at room temperature of 32°C. during summer did not germinate.

When the spore germinates, the epispore ruptures before the promycelium comes out into which the protoplasm of the spore passes. The promycelium varies in length from 10 μ or less to something like 1·5 mm. (1,500 μ) or more. Generally it is unbranched but sometimes branched promycelium has also been noticed. The promycelium has a tendency to go on elongating and as it elongates the basal portion degenerates. The contents of the promycelium are

coarsely granular and the protoplasm moves continuously forward, so that the protoplasm is confined to the upper portion, the portion next to spore becomes vacuolated, and later entirely vacant when septa are laid down. At the tip of the promycelium a crown of slender sporidia is formed in a whorl. The formation of sporidia is often delayed on account of the continuous growth of the promycelium. The number of sporidia varies from 32 to 128 and even more. (Plate VI, figs. 2 and 3 and Plate VII figs. *d*—*h*). Fusion of two sporidia has never been noticed. Spores germinating in hanging drop cultures or on solid media were kept for more than a month but no fusion of sporidia was noticed and no further growth took place.

VII. Infection experiments carried out at Pusa and Karnal

It has already been mentioned that *T. indica* does not occur nor has so far been recorded from places outside North-West India. The writer has been making careful observations for this bunt for the last four years in Bihar but has not come across a single bunted grain in any grain market of that province. Hundreds of maunds of wheat were carefully examined every year in the Botanical Section and the Farm at Pusa but no trace could be obtained of this bunt. Even seed from Karnal sown at Pusa never showed any sign of infection. It was also observed that partly bunted grain of *T. indica* does germinate and produce seedlings. In order to verify the observations made and to see whether it is possible to produce bunt in Pusa under ordinary climatic conditions by sowing naturally infected seeds at the normal sowing time or by sowing late when the temperature has cooled down, the following experiments were carried out in soil, in pots and in plots.

Experiment I.—Bunted grains of five wheat varieties collected during May and June 1930 were sown in Pusa in sterilized soil in pots in November. There were ten pots for each variety and each pot had six seedlings, that is, altogether fifty pots and 300 seedlings. At maturity during April 1931, all the plants formed normal ear and produced healthy grains. Seeds of the same varieties kept under similar conditions when sown in Karnal produced bunt.

Experiment II.—A parallel experiment was carried out at Pusa in which diseased seeds obtained from Karnal were given an extra dose of artificial infection by mixing the seed with spore powder. The spore powder was obtained at the time of infection by crushing naturally infected grain kept under dry conditions. None of the ears of the 300 wheat plants produced a single bunted grain.

Experiment III.—In order to see if sowing in the cold weather or when the climatic conditions are somewhat like those of Karnal help in producing bunt at Pusa, diseased seeds of Pusa 113 collected during May 1933 at Karnal were sown at different intervals during November and December of the same year at Pusa and the details are given in Table IV.

TABLE IV

Infection experiment in sterilized soil in pots with bunted grain during 1933-34

Date of sowing	No. of pots	No. of plants	Infection
2nd November 1933	20	100	*Nil*
20th „ „	20	100	„
29th „ „	20	100	„
6th December „	20	100	„

The crop was harvested in April 1934 and each and every ear was threshed by hand and every grain was carefully examined but not a single bunted grain was noticed. The seeds of this same wheat type kept under similar conditions when sown in Karnal had three per cent bunted ears.

Experiment IV.—The diseased seeds of four wheat varieties collected at Karnal during May and June 1933 were sown in a plot at Pusa on 5th December 1933, that is, five or six weeks after the normal time of sowing. The idea of sowing late was to secure as far as possible climatic conditions prevailing at Karnal at the sowing time. Each variety was sown in several plots. Pusa 113 seed from the Botanical Section, Pusa, was also sown in a contiguous plot. The number of ears in all the plots were counted and final observations were made at the time of maturity. The details are given in Table V.

TABLE V

Infection experiment with bunted wheat grain during 1933-34 at Pusa under field conditions

Wheat variety	Replications	No. of ears	No. of bunted ears	Remarks
Pusa 113	1	894	*Nil*	Seed from Botanical Section, Pusa & free from bunt
	2	710	,,	
	3	640	,,	
	4	638	,,	
Pusa 113	1	620	,,	Seed from Karnal & heavily infected with bunt
	2	596	,,	
	3	651	,,	
	4	1,189	,,	
Pusa 113	1	423	,,	Selected heavily infected seed from Karnal
	2	618	,,	
Pusa 4	1	331	,,	Ditto.
	2	493	,,	
	3	452	,,	
	4	559	,,	
	5	577	,,	
Pusa 72-1-1	1	498	,,	Seed from Karnal and heavily infected with bunt
	2	567	,,	
	3	523	,,	
	4	649	,,	
Pusa 165	1	462	,,	Selected heavily infected seed from Karnal
	2	586	,,	
	3	490	,,	
	4	436	,,	
	5	436	,,	

At the time of harvesting at the end of April 1934 each and every head was examined and threshing was done by hand but not a single infected grain was noticed.

Experiment V.—During 1932-33 bunt was up to twenty per cent on Pusa 72-1-1 at Karnal. Part of this seed was brought to Pusa and sown as shown in Table V. The total number of ears obtained in Pusa from four plots in which this variety was sown was 2,237 and none produced bunted grain. The seed from the same lot was also sown in Karnal in twelve plots and the total number of ears counted was 8,535, out of which 455 ears produced bunted grain giving 5·33 per cent of infected ears. The details are given below in Table VI.

TABLE VI

Infected seed of Pusa 72-1-1 sown at Karnal

Replications	No. of ears	No. of ears bunted	Percentage
1	584	28	4·79
2	670	32	4·77
3	695	34	4·89
4	765	36	4·70
5	870	42	4·82
6	670	33	4·92
7	618	36	4·82
8	750	44	5·86
9	895	53	5·92
10	611	35	5·72
11	720	42	5·83
12	686	40	5·83
Total	8,535	455	5·33

Experiment VI.—An experiment similar to Experiment V was carried out at Karnal using Pusa 113 from the same lot from which it was used in Experiment IV and shown in Table V. The total number of ears produced at Pusa was 2,956 from four plots and none produced bunted grain whereas at Karnal this type was sown in five plots and the total number of ears counted was 4,767, out of which 50 ears produced bunted grain giving 1·1 per cent of infected ears.

Seeds of Pusa 165 which were brought to Pusa from Karnal and sown in five plots had no bunted grain whereas same seed at Karnal produced a fair amount of bunt.

Thus the seeds of Pusa 72--1--1, 113 and 165 which were stocked at Karnal and brought to Pusa at the time of sowing produced bunt at Karnal and not at Pusa. This shows clearly that climatic conditions at Pusa are responsible for the failure of infection.

Experiment VII.—In addition to the above experiments a very large number of diseased seeds were kept in an incubator at 22°C. and when germinated were sown partly in the end of November and partly early in December in pots.

Seedlings when four to six inches in height were taken out and microscopically examined. In several of them mycelium similar to that of a bunt was noticed especially at the nodes. No mycelium was found in grown up plants. The plants left after examination and allowed to form ear produced no bunt.

Experiment VIII.—In order to see whether the bunt is in a dormant state on wheat in Pusa, that is, whether it will produce infection if favourable conditions are given, seeds of Pusa 111, 112 and 113 from the Botanical Section, Pusa, were taken to Karnal and sown side by side with Karnal seed. Each variety was sown in twelve plots and each plot had on an average 650 ears. Minute examination of individual grain revealed total absence of bunt. The crop was hand-threshed and grains of all the thirty-six plots were very carefully examined but no bunt was noticed.

Pusa 113 (Karnal seed) was also sown side by side in twenty-four plots and showed approximately 2·5 per cent of bunted ears. This shows that Pusa seed is free from the disease.

The fact that the disease has never been observed at Pusa or in other parts of Bihar, and the non-occurrence of the disease when Pusa seed is sown at Karnal or when Karnal seed is sown at Pusa support the view that the climatic conditions prevailing in Bihar are unsuitable for the spread of this disease. Further, seeds kept under identical conditions were able to produce bunted grain at Karnal and not at Pusa. Late sowing when the temperature lowers also does not help the bunt to appear at Pusa. By late sowing the crop is harvested late and it appears that high temperature at the time of maturity is unfavourable for the bunt to grow in the tissue of the host plant.

VIII. Experiments on control measures

In order to determine the effect of some fungicides on the incidence of *T indica* an experiment was carried out at the Botanisal Sub-Station, Karnal, in 1933-34. The fungicides tested were copper carbonate, ceresan, formalin and uspulun (universal). Untreated wheat seed was also sown as control, thus making up five treatments in the experiment. Three varieties of wheat used were Karnal-grown seed of Pusa 111, 112 and 113. There were two methods of infection, *viz.*—

Series A. Naturally infected seed

Series B. Naturally infected seed plus an extra dose of artificial infection by shaking every 100 grms. of seed with one grm. of bunt powder

The treatments were as follows in both the series:—

(i) Control—untreated seed—naturally infected in series A and a further dose of artificial infection in series B as stated above

(ii) Seed treated with finely powdered copper carbonate at the rate of two ounces per bushel (60 lbs.)

(iii) Seed treated with ceresan at the rate of two ounces per bushel

(iv) Seed treated with formalin solution 0·25 per cent for ten minutes and exposed to formalin vapour for four hours.

(v) Seed treated with uspulun (universal) 0·25 per cent solution for thirty minutes

The experiment was carried out on a piece of land which was laid out on a randomized block system with eight replications. Each block contained thirty plots, each 10 ft. × 5 ft. in size, fifteen under infection series A and fifteen under infection series B. Each plot had five rows of plants. There were five treatments and three varieties in each series.

At the time of harvest all the ears in each plot were counted and the number of ears showing bunted grain was noted down. The total number of healthy and bunted ears in each of the eight blocks under both series of treatments for the three wheat varieties separately is shown in Table VII. The amount of bunt in each plot is given as a percentage of the total number of ears, which is shown in Table VIII.

Table VII

The number of healthy and bunted ears in three varieties of wheat

Treatment	Varieties	Block 1				Block 2				Block 3				Block 4				Block 5				Block 6				Block 7				Block 8			
		A		B		A		B		A		B		A		B		A		B		A		B		A		B		A		B	
		H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D	H	D
Control . .	P 111	530	5	608	45	750	6	550	21	701	6	461	16	825	7	453	17	485	8	390	23	702	4	812	70	720	5	640	21	836	4	463	14
	P 112	517	8	620	31	598	10	636	37	450	24	523	51	550	16	490	50	520	10	611	50	750	20	750	41	640	25	811	27	750	16	640	40
	P 113	480	16	714	16	552	16	570	13	566	18	678	24	650	20	635	16	564	16	605	29	820	11	593	12	805	20	550	16	665	16	720	23
Copper carbonate	P 111	508	2	312	1	511	1	550	1	480	2	586	1	365	1	680	2	380	1	732	3	704	3	850	2	653	8	612	2	324	5	416	2
	P 112	360	2	500	4	572	10	580	2	405	4	462	2	570	5	675	3	557	5	795	3	840	4	839	1	680	4	574	2	535	3	654	4
	P 113	770	1	548	4	587	2	710	2	710	1	722	2	665	0	670	4	540	1	610	4	872	2	1105	1	662	1	600	1	710	1	824	5
Ceresan	P 111	525	1	454	2	440	3	380	3	565	2	716	2	412	1	735	3	549	1	850	3	808	2	865	3	542	3	350	2	610	1	402	2
	P 112	435	4	520	5	521	4	710	6	413	8	550	6	634	10	800	4	542	7	1107	13	725	6	814	2	536	5	695	6	654	9	928	3
	P 113	436	1	480	1	510	2	782	4	532	2	840	1	480	2	690	1	424	1	518	1	654	1	809	2	420	2	530	2	440	1	701	3
Formalin	P 111	450	10	60	0	900	6	84	0	360	4	125	0	370	6	153	0	593	5	224	2	450	4	329	2	320	4	45	0	430	6	51	0
	P 112	460	3	210	1	459	5	600	2	572	4	191	0	470	3	240	6	583	5	360	5	581	5	520	8	620	2	200	2	690	5	158	5
	P 113	708	3	308	1	625	3	365	1	605	5	215	2	531	6	225	1	672	3	302	3	990	5	470	5	470	1	324	0	714	1	225	3
Uspulun (Universal)	P 111	622	1	453	1	520	1	240	1	653	1	540	3	470	0	504	1	850	1	550	0	826	1	650	4	605	2	575	2	471	0	460	1
	P 112	480	2	519	0	620	1	750	2	570	1	667	2	760	6	732	3	736	2	800	4	410	2	870	2	570	6	820	1	527	1	590	2
	P 113	525	7	540	0	480	2	710	6	810	2	645	1	940	2	716	3	830	4	805	5	915	3	1010	8	520	3	480	3	450	2	716	2

H—Total No. of healthy ears ; D—Total No. of bunted cars. A—Series A. B.—Series B.

TABLE VIII

Percentage of infection

Block	Varieties / Infection series	Treatment: Control P111	Control P113	Control Pl 13	Copper carbonate P111	Copper carbonate P112	Copper carbonate P113	Ceresan P111	Ceresan P112	Ceresan P113	Formalin P111	Formalin P112	Formalin P113	Uspulun P111	Uspulun P112	Uspulun P113	Infection total Series A	Infection total Series B	Block totals A+B
1	A	·94	1·54	3·33	·39	·55	·12	·19	·19	·22	2·22	·65	·43	·16	·41	1·33	13·38	—	
	B	7·40	5·00	2·23	·32	80	·72	·44	96	·20	·00	·47	·32	·23	·00	·00	—	19·08	32·46
2	A	·80	1·67	2·89	·19	1·74	·34	·68	·76	·39	·66	1·08	·48	·19	·16	·41	12·44	—	
	B	3·81	5·81	2·28	·18	·34	·28	·78	·84	·51	·00	33	·27	·41	·28	·84	—	16·96	29·40
3	A	·85	5 33	3·18	·41	·98	·14	·35	1·93	·37	1·11	·69	·82	·15	·17	·24	16·72	—	
	B	3·47	9·75	3·53	·17	·43	·27	·27	1·09	·11	·00	·00	·93	·55	·29	·05	—	21·01	37·73
4	A	·84	2·90	3·07	·29	·87	·00	·24	1 57	·41	1·62	·63	1·12	·00	·78	·21	14·55	—	
	B	3·75	10·20	2·44	·29	·44	·59	·40	·50	·14	·00	2·50	·44	·19	·40	·41	—	22 69	37·24
5	A	1·64	1·92	2·83	·26	·89	·18	·18	1·22	·23	·84	·85	·44	·11	·27	·48	12·34	—	
	B	5 89	8·18	4·79	·46	·37	·65	·55	1·17	·19	·89	1·35	·99	·00	·50	·62	—	26·37	38·71
6	A	·56	2·66	1·34	·42	·47	·22	·24	·82	·15	·88	·86	·50	·12	·48	·32	10·04	—	
	B	8·62	5 48	2·02	·23	·11	·09	·34	·24	·24	·60	1·53	1·06	·61	·23	·79	—	22·16	32·20
7	A	·69	3·90	2·48	1·22	·58	·15	·55	·93	·47	1·25	·32	·21	·33	1·05	·57	14·70	—	
	B	3·28	3·33	2 90	·32	·34	·16	·57	·86	·37	·00	1·00	·00	·34	·12	·62	—	14·20	28·90
8	A	·47	2·13	2·40	1·54	·56	·14	·16	1·37	·22	1·39	·72	1·40	·00	·18	·44	13·12	—	
	B	3·02	6·25	3·19	·48	·61	·60	·49	·32	·42	·00	3·16	1·33	·21	·33	·27	—	20·68	33·80
Varietal series total	A	6·79	22·05	21·52	4 72	6 64	1·29	2·59	9·51	2 46	9·97	5·80	5·39	1·06	3·50	4·00	107·29	—	
,, ,,	B	39·24	53·07	23 38	2·39	3·44	3 36	3 64	5·98	2 ..8	1·49	10·37	5·34	2·53	2 14	3·70	—	163·15	270·44
Varietal totals of A+B		46·03	76·02	44·90	7·11	10·08	4·65	6·23	15 49	4·64	11·46	16·17	10·73	3 59	5·64	7·70	107·29	163·15	270·44
Treatment totals		166·95			21·84			26·36			38·36			16·93			270·44		270·44

It will be noted from the tables that infection in series B is heavier than that in series A, that is, the extra dose of infection given in series B has definitely increased the percentage of bunt. It will also be observed that uspulun (universal) though unable to control the disease altogether gave the least percentage of bunted ears.

Statistical analysis of the data is worked out according to Fisher's analysis of variance [1932] as shown in Table IX.

TABLE IX

Analysis of variance

Variance due to	Degrees of freedom	Sum of squares	Mean square	Observed value of Mahalanobis' x	Critical value of Mahalanobis' x = ·01)
Block	7	3.305	0.472	..	..
Treatments . . .	4	336.974	84.244	155.144	3.320
(Control and fungicides) Infections (A and B) . .	1	13.002	13.002	23.946	6.635
Varieties (Pusa 111, 112 and 113)	2	20.755	10.378	19.111	4.605
Interactions . . .	22	149.881	6.813	12.547	1.791
Residual	203	110.239	0.543	..	..
Total . .	239	634.156	..	..	..

It will be seen from the above table that the calculated value of x exceeds the theoretical value given in Mahalanobis' table [1932] for one per cent level of significance in all items except blocks showing thereby that the differences between the several treatments, infections and varieties are statistically significant. The differences between the mean values of the different treatments—control, copper carbonate, ceresan, formalin and uspulun (universal) are given in Table X.

TABLE X

The differences between the mean values for treatment

Treatments	1 Control	2 Copper carbonate	3 Ceresan	4 Formalin	5 Uspulun
Control	..	—3.023	—2.929	—2.679	—3.125
Copper carbonate . .	+3.023	..	+0.094	+0.344	—0.102
Ceresan	+2.929	—0.094	..	0.250	—0.196
Formalin	+2.679	—0.344	—0.250	..	—0.446
Uspulun	+3.125	+0.102	+0.196	+0.446	..
Mean	3.478	0.455	0.549	0.799	0.353

The critical differences for treatments have been calculated and found to be:—

Critical difference = 0·304 (P = ·5)

Critical difference = 0.401 (P = ·01)

A comparison of the various figures with the critical differences leads to the following conclusions:—

Treatment 5 (uspulun universal) is superior to treatment 4 (formalin) at the one per cent level. Treatment 2 (copper carbonate) is superior to treatment 4 (formalin) at the five per cent level. Treatment 2 (copper carbonate), treatment 3 (ceresan), treatment 4 (formalin) and treatment 5 (uspulun) are superior to treatment 1 (control—undisinfected seed) at the one per cent level. There is no significant difference between other combinations.

The differences between the mean of different varieties *viz.* Pusa 111, 112 and 113 are given in Table XI.

TABLE XI

The differences between the mean values for varieties

Varieties	Pusa 111	Pusa 112	Pusa 113
Pusa 111	..	0.613	—0.022
Pusa 112	—0.613	..	—0.535
Pusa 113	0.022	0.535	..
Mean	0.930	1.543	0.908

Critical difference = .229 (P = .05)

Critical difference = 0.303 (P = ·01)

From the above table it is evident that Pusa 112 is inferior to Pusa 111 = Pusa 113 at the one per cent level in respect of susceptibility to bunt

Comparing the two series of infections we find that

Mean of A = 0·893

Mean of B = 1·360

∴ Mean difference = 0·467

Critical difference = 0.247 (P = ·01)

Thus infection is significantly higher in the B series than in the A series as was expected on account of extra dose of infection given in the former.

In another experiment heavily infected seed of Pusa 72-1-1 was treated and sown in Karnal under the following treatment:—

1. Control—heavily infected seed
2. Seed treated with copper carbonate at the rate of two ounces per bushel
3. Seed treated with ceresan at the rate of two ounces per bushel

At the time of harvesting all the ears were counted and those having bunted grain were also noted. The following data give the percentage of bunted ears.

TABLE XII

The effect of various fungicides on T. indica.

Treatment	No. of ears	No. of ears with bunted grain	Percentage of bunted ears
Control	8535	455	5.33
Copper carbonate	4105	68	1.65
Ceresan	4152	1081	2.60

This experiment also shows that copper carbonate and ceresan are able to reduce the percentage of bunted ears from 5.33 to 1·65 and 2·60 respectively but none of these treatments can completely control the disease. Untreated seed of this variety which produced 5.33 per cent of bunted ear under Karnal conditions was sown at Pusa as explained in Experiment V but produced a healthy crop with no bunted ear.

Microscopic examination of lightly diseased grain shows that the fungus occurs under the seed-coat and that it is well protected by the pericarp, especially in the lightly attacked grain. The fungicides therefore are unable to kill the internal mycelium or the spores, though they are able to reduce the percentage of disease. Hot water treatment may be a possible method to check the disease to a greater extent which will, in addition destroy loose smut (*Ustilago tritici* (Pers.) Jens.). Luthra [1934] has found that dipping the seed in water for four hours and then exposing it to solar heat for four or five hours during summer can check the loose smut. It remains to be seen whether this treatment will check bunt as well. Experiments are being conducted to see the efficacy of the hot water treatment for bunt control and should the method be successful, it will be very useful in controlling both bunt and loose smut by one and the same treatment.

During 1932-33 bunt was very slight or almost absent on Pusa 114 at Karnal and during 1933-34 seed of Pusa 114 treated with ceresan was sown in a plot in which during 1932-33 there was Pusa 111 showing bunt upto an extent of two per cent. An examination of seed of Pusa 114 showed a fair amount of bunt. It appears that the soil was infected from bunt spores fallen from Pusa 111 during 1932-33 and this perhaps may have infected Pusa 114. That infection takes place from soil in other species of Tilletia is well known. It is quite possible that Pusa 114 got infection from the soil infected previously. Thus rotation of crops is also advisable and wheat should not, if possible, be sown on land occupied in the previous year by the same crop.

IX. Conclusion and summary

Bunt on wheat in India is caused by three species of Tilletia, that is, *T. caries*, *T. foetens* and *T. indica* and is confined to the north western parts of India and does a fair amount of damage. *T. caries* and *T. foetens* are restricted to the cooler regions while *T. indica* is confined to the plains. All the three species possess a stinking smell. *T. indica* can very easily be distinguished from other species by its partial attack on the grain and also by its black coloured spore mass, while in the other two species the whole grain except the seed-coat is destroyed and the spores are dusty olive brown or rust coloured in mass. Spores of *T. indica* are much larger than those of *T. caries* to which it is closely allied and it appears that there are at least two physiologic forms of *T. indica*. The biometric analysis of data of spore measurement shows that this method can be employed to determine physiologic forms though it is not enough to identify the different species. There is little possibility of any wind dissemination of spores directly from the bunted head as occurs in the case of *Ustilago tritici* (Pers.) Jens. The bunted grain is wholly or partly concealed by the glumes and before the harvest there is little or no opportunity for the dissemination of the bunt spores. When the crop is mature, the bunted grain is sometimes easily displaced and falls on the ground with a slight disturbance and thus infects the soil. The danger of dissemination of bunt spores lies in the threshing operation when they adhere to the surface of the sound grain and lodge in the brushes. Large number of grains thus get infected. When the bunted grains come in contact with the healthy ones in storage, they infect the latter very easily. It has also been shown by a series of infection experiments done at Karnal and Pusa that infection does not take place in Pusa whereas at Karnal the disease appears almost every year. It is due to different climatic conditions.

Control measures tried show that the percentage of infection can be reduced by treatment with fungicides like uspulun (universal), copper carbonate, ceresan and formalin but none of them is able to check the disease altogether, the reason being that spores are well protected in mildly attacked grain by the pericarp and the fungicide cannot reach the spores. It is supposed that hot water treatment may be a possible method to check the disease to a greater extent, and as loose smut is also very common on wheat, one treatment might do for both the diseases. Further, there is an indication that as in other bunts infection can take place from infected soil and so rotation of crops is advisable. It would be better if from time to time Karnal seed is renewed with seed from Pusa which is free from both bunt and smut.

Acknowledgments

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TILLETIA INDICA

Infected wheat ears. Two spikelets with bunted grain and a heavily infected grain are also shown.

TILLETIA INDICA

Infected wheat grain with various stages of infection from mild to severe.

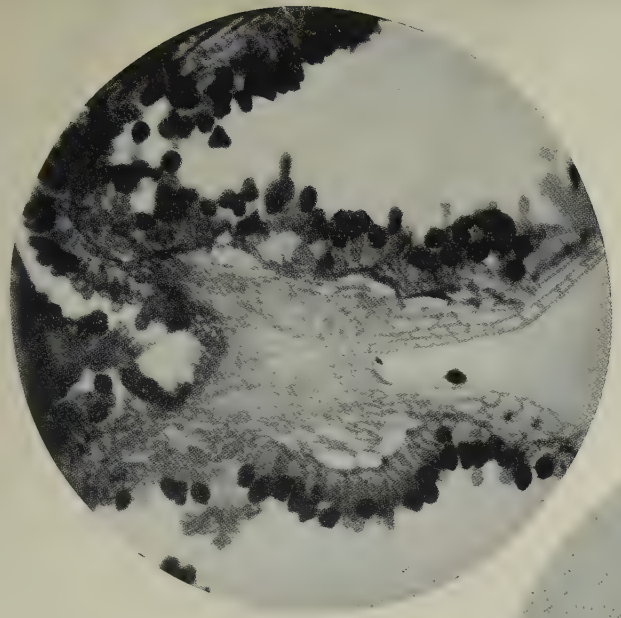


FIG. 1.

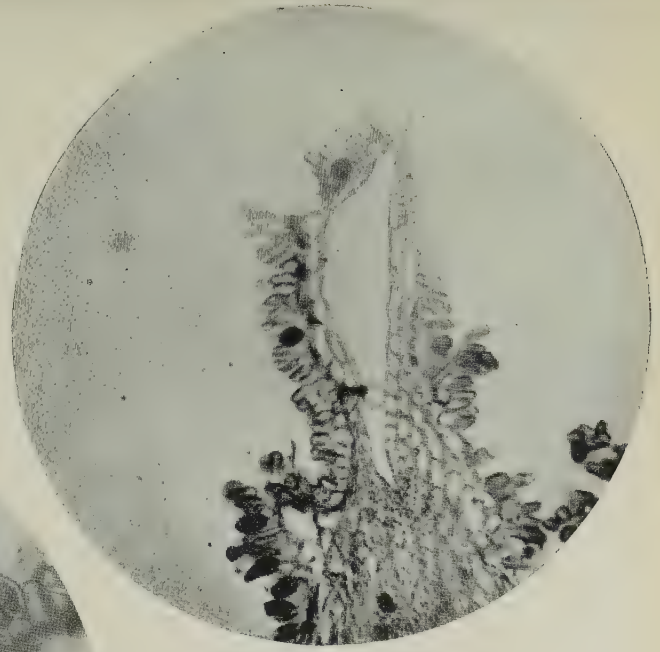


FIG. 2.

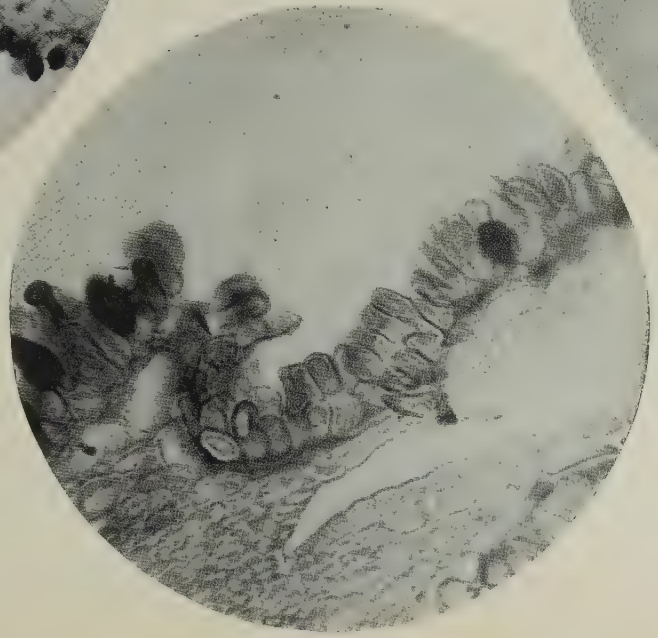


FIG. 3.

Photomicrographs of sections of infected grain showing sori and formation of spores (×500).

TILLETIA INDICA

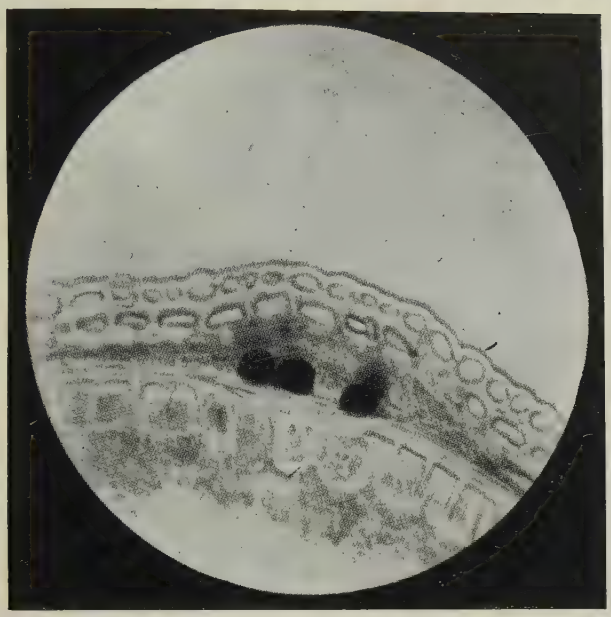


FIG. 1.

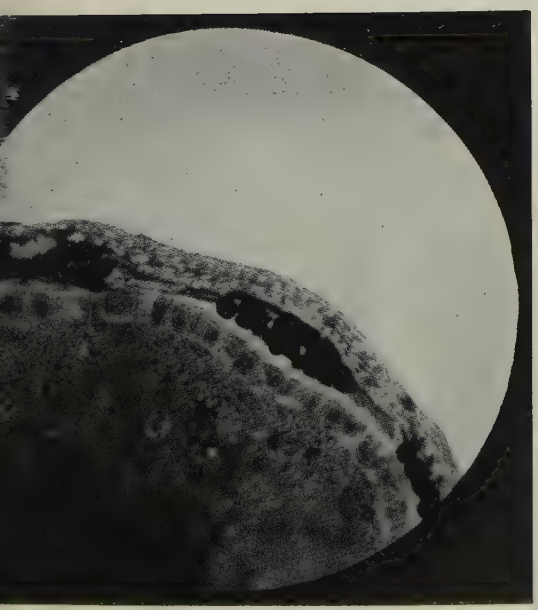


FIG. 2.

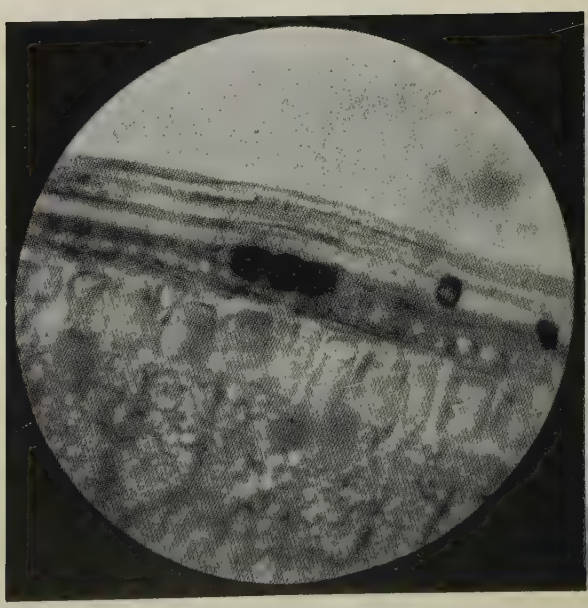


FIG. 3.

Spore formation in lightly infected grain under the seed-coat and above the aleurone layer of dosperm (×500).

TILLETIA INDICA

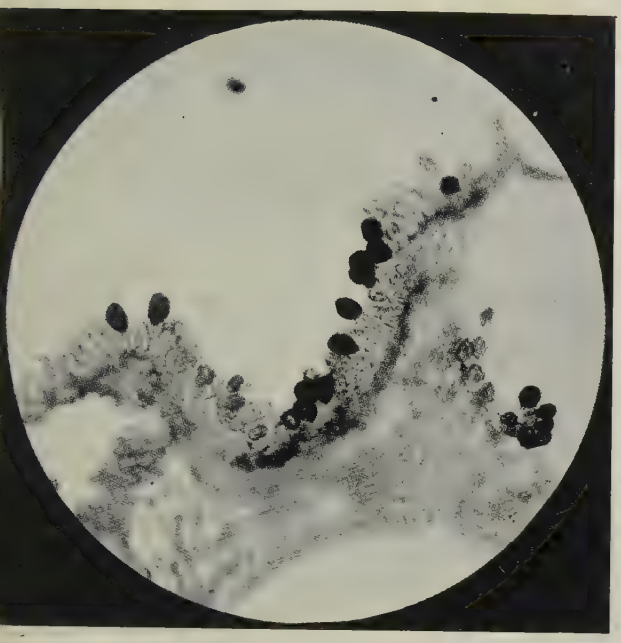


FIG. 1. Spores and sterile cells in sori.

FIG. 2. Formation of a sorus in the tissue on the ventral surface.

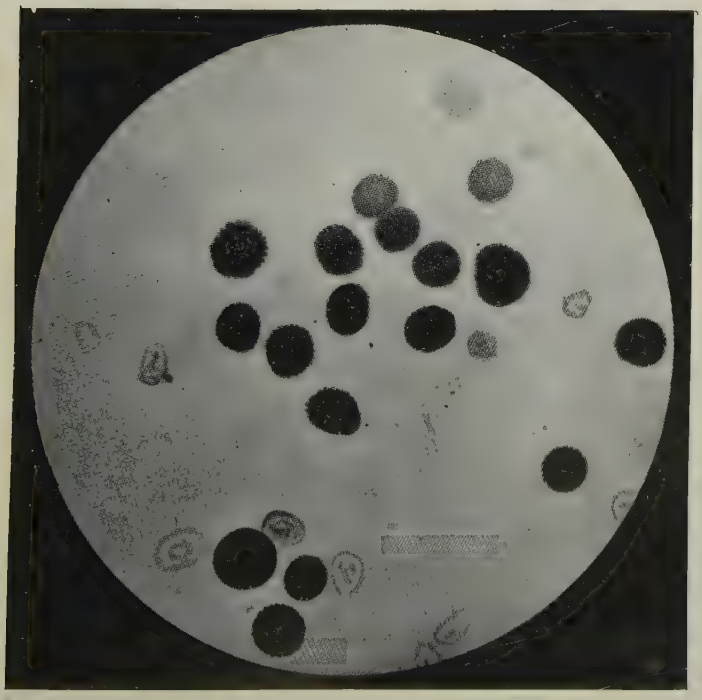


FIG. 3. Mature and young spores with lighter colour sterile cells.

TILLETIA INDICA

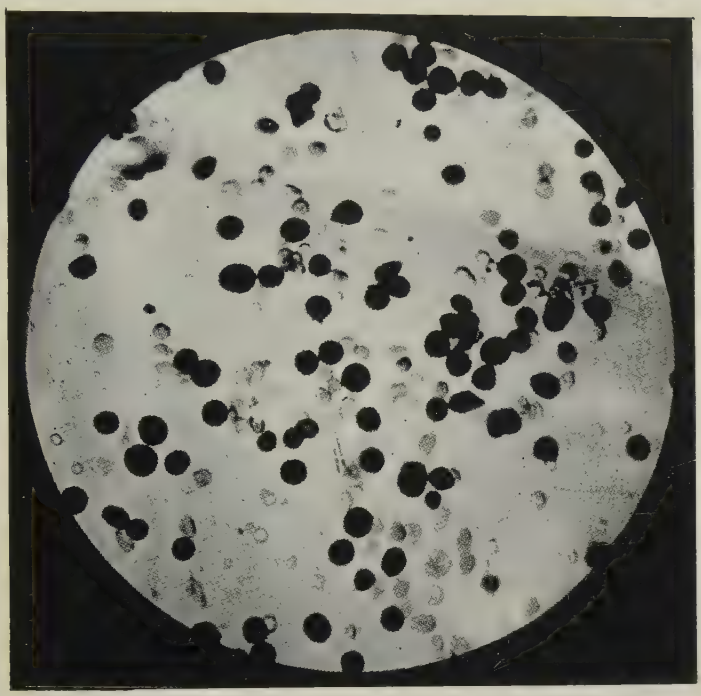
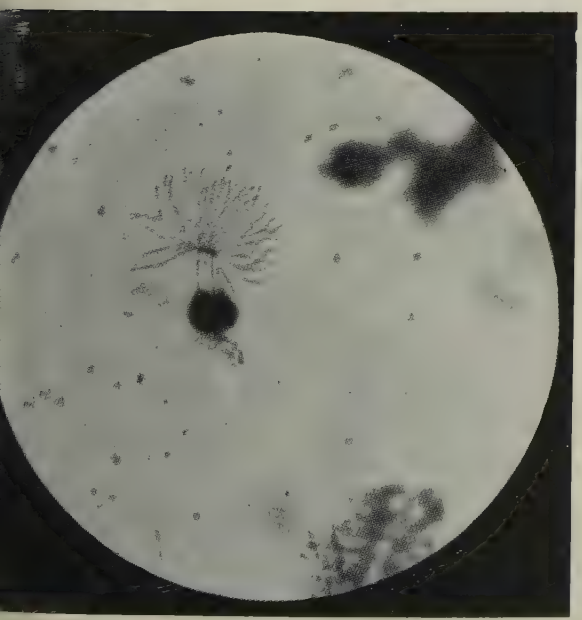


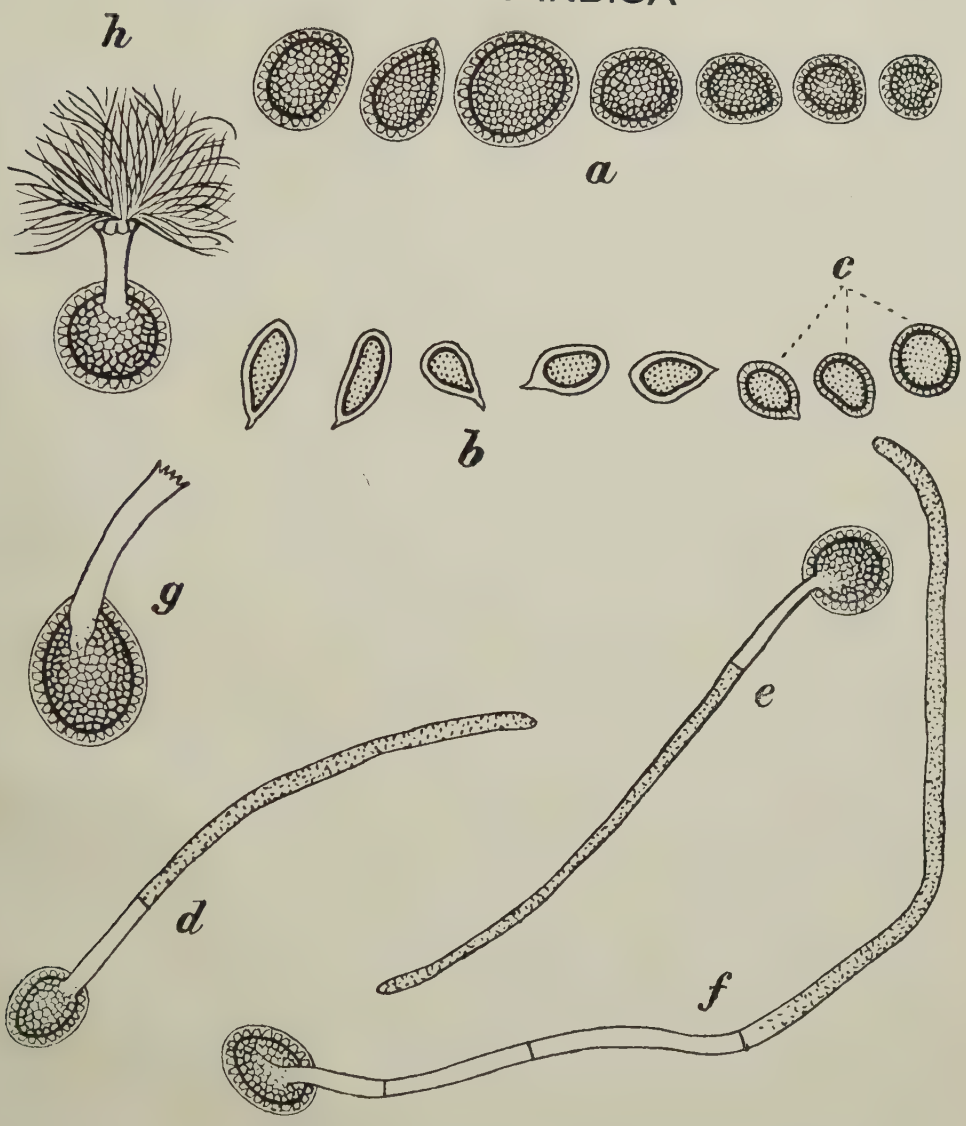
FIG. 1. Mature and young spores with lighter colour sterile cells.



2. A germinated spore with a short promycelium bearing a crown of sporidia.

FIG. 3. A germinating spore with a promycelium.

TILLETIA INDICA



(a) Mature spores with reticulation of epispore. One spore with a short papilla (×200).

(b) Sterile cells (×200).

(c) Young spores. One with a papilla (×200).

(d), (e) & (f) Germination and formation of promycelium (×200).

(g) A germinated spore showing the place of attachment of sporodia (×200).

(h) A germinated spore bearing a crown of sporodia (×200).

THE RELATION OF SOME PLANT CHARACTERS TO YIELD IN SORGHUM

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The sorghum plant has been under study at the Millets Breeding Station for some years. A number of characters of a qualitative kind have been pursued and in May 1931 a study of the relationship between some of the prominent plant characters and the all-important question of yield was commenced. A rough summary of the results of first attempts was given [Rangaswami Ayyangar, 1932] in the 'Symposium on Sorghum' on the occasion of the Annual Conference of the Madras Agricultural Students' Union in December 1931.

Materials and methods

These studies have been made in three seasons, two of them summer (irrigated) and one main (rain-fed). Five Coimbatore varieties were the subjects of study. They are listed below:—

Season	Strain No.	Name of variety	Colour of grain	Year	Number of plants measured
Summer (irrigated) . .	*A. S.* 809	*Chinna Manjal*	Yellow .	1931	240
				1932	124
	„ 1063	*Uppam Cholam*	White	1931	243
				1932	125
Main (rain-fed) . . .	„ 60	*Peria Manjal*	Yellow (Brown wash).	1931	250
	„ 1098	„	„	1931	250
	„ 129	*Sen Cholam*	Red .	1931	250

All the varieties belong to the group *Sorghum durra* Stapf. The sorghum plant being single-eared the yield is not complicated by tillers as in other cereals. The individual plants were taken from a bulk plot, care being taken to steer clear of diseased individuals and plants at borders. At 'shot blade' stage, plants were labelled and numbered in the field.

The following characters were chosen:—1. Grain yield. 2. Thickness of earhead, 3. Length of earhead, 4. Diameter of peduncle, 5. Weight of dry straw, 6. Weight of 100 grains, 7. Weight of earhead—fresh, 8. Weight of earhead—dry, 9. Length of peduncle, 10. Duration.

The relation of yield to the other characters is, in this study, treated by the method of correlation. The suffix of r is as numbered in the list of characters enumerated above. In Table I are presented the total correlations of grain yield with eight characters (and one more character, *i.e.*, duration, in one season).

TABLE I

Total correlation coefficients

Character correlated with grain yield	Irrigated				Rain-fed 1931 Main		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
Thickness of earhead	.772	.813	.851	.712	.787	.768	.781
Length of earhead	.748	.535	.701	.457	.828	.775	.847
Diameter of peduncle	.845	.739	.879	.569	.870	.855	.857
Weight of dry straw	.588	.451	.702	.375	.763	.828	.835
Weight of 100 grains	.701	.805	.713	.731	.260	.394	.175
Weight of earhead—fresh.	.927	.976	.986	.954	.968	.960	.979
Weight of earhead—dry	.982	.972	.992	.953	.984	.999	.982
Length of peduncle	—.253	—.559	—.097	.119	.203	.157	—.137
Duration	—.685	..	—.601	..	..	..	..
Level of significance *P*.01	.165	.227	.165	.227	.161	.161	.161

As there are seven coefficients for each pair of factors, it is expedient to test whether the same degree of correlation is shown (1) by the same variety in two seasons, (2) in the same season by two varieties, (3) by the irrigated and the rain-fed varieties by themselves, and (4) by all the seven taken together. The test is applied after the hyperbolic transformation $z = \tanh^{-1} r = \frac{1}{2}\{\log_e (1+r) - \log_e (1-r)\}$. In columns 2 to 5 of Table II, the significance of the difference of the z's is measured by considering whether $\frac{Z-Z'}{\sigma_{z-z'}}$ is greater or less than $2\ \chi^2 = S(z-\bar{z})^2 (n-3)$ where $\bar{z}$ is the weighted mean of the z's, (the weights being proportional to n-3) and n, the size of the sample. χ_1^2 and χ_2^2 in columns 6 and 7 relate to irrigated and rain-fed varieties respectively; χ^2 combines all the seven coefficients. Further it can easily be shown that χ^2 is greater than $\chi_1^2 + \chi_2^2$. Therefore, if $\chi_1^2 + \chi_2^2$ exceeds the χ^2 level for $P = \cdot 05$, χ^2 need not be calculated separately. For the details of the technique employed, reference may be made to Tippett's "The Methods of Statistics" [1931]

TABLE II

Combination of the correlation coefficients

Character correlated with grain yield	$\frac{z-z^1}{s_z-z^1}$				χ_1^2 Irrigated $\chi_1^2=7\cdot82$ $P=\cdot05$	χ_2^2 Rain-fed $\chi_2^2=5\cdot99$ $P=\cdot05$	χ^2 $\chi^2=12\cdot59$ $P=\cdot05$
	A. S. 809 Summer 1931 and 1932	*A. S.* 1063 Summer 1931 and 1932	Summer 1931 *A. S.* 809 & *A. S.* 1063	Summer 1932 *A. S.* 809 & *A. S.* 1063			
(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
Thickness of earhead .	1·00	3·36	2·53	1·95	12·96	·22	..
Length of earhead .	3·36	3·45	1·10	0·86	29·99	4·40	..
Diameter of peduncle .	2·64	6·55	1·43	2·34	48·72	0·42	..
Weight of dry straw .	1·73	4·36	2·17	0·70	23·38	60·02	..
Weight of 100 grains .	2·18	0·36	0·22	1·64	5·16	7·26	162
Weight of earhead—fresh	5·18	5·55	9·23	2·66	91·27	13·06	..
Weight of earhead—dry	2·00	8·18	4·51	2·11	72·86	>1000	..
Length of peduncle .	3·36	2·00	1·76	5·86	26·75	18·48	..
Duration . . .	..	..	1·56	..	..	..	..

The correlations show a great deal of variation from variety to variety and season to season; this is apparent from columns (2) to (5) of Table II. From column (6) it is evident that in the irrigated varieties, the same degree of correlation exists between grain yield and the weight of 100 grains. In the case of rain-fed varieties, the correlation coefficients for the several varieties for the thickness of earhead are not different; the same can be said in the case of the length of earhead and diameter of peduncle. But when the coefficients for the several varieties are considered together, they show wide range of variability.

That the correlations show a great deal of variation is not surprising. The relation of one character with another, in the complex plant life, is intricately dependent upon a group of other factors tangible and intangible, and changes in these factors caused by varietal and seasonal fluctuations, will be reflected in the original correlations measured. In the present study, wherever the regressions are not far from linearity, partial correlations are employed to estimate the correlations eliminating some characters. Multiple correlations have also been computed to find whether a knowledge of additional factors helps in the prediction of yield.

Instead of the probable error, the level of significance for $P = \cdot 01$ is calculated for every correlation coefficient [Fisher, 1930]*; this is helpful in judging the significance of the values at sight.

Relation between earhead length and thickness, and yield.—The earhead consists of a group of spikes disposed in whorls round a stalk. This stalk varies in length. In the economic varieties of grain sorghums it stops short of reaching the very top of the earhead, and from its apex radiate spikes which fill and give the earhead the full and globular look it has; so that for the purpose of convenient and practical measurement, the length from the base to the tip of the earhead as a whole was preferred to that of merely the rachis.

The thickness of the earhead was measured by placing it across a metre scale and bringing together two rectangular pieces of wood so as to touch the head on either side without pressing it and the distance between the inner faces of the blocks read direct from the scale. This gives the maximum thickness of the panicle. In the varieties studied this maximum thickness was observed to be at about a third of its length from the base.

The earhead thickness and length are highly correlated with yield in all the varieties and in all the seasons. This is as it should be, considering that the length and thickness are the two dimensions determining the volume of the earhead.

---

* The value of the correlation coefficient for the ·01 level of significance is taken as $\frac{t}{\sqrt{n - 2 + t^2}}$, where n is the number of pairs of observations, and considering that n is large, t is taken as 2·576 corresponding to $n = \infty$.

TABLE III

Partial correlations

Grain yield and thickness of earhead

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	A. S. 1098	*A. S.* 129
r_{12} . . .	·772	·813	·851	·712	·787	·768	·781
Level of significance ($P = ·01$).	·165	·227	·164	·226	·161	·161	·161
$r_{12·3}$ . . .	·509	·725	·863	·622	·275	·469	·626
$r_{12·4}$ . . .	·351	·532	·493	·523	·356	·528	·645
$r_{12·5}$ . . .	·643	·779	·716	·667	·612	·589	·703
$r_{12·6}$ . . .	·694	·738	·765	·714	·788	·757	·780
Level of significance ($P = ·01$).	·165	·228	·164	·227	·162	·162	·162
$r_{12·34}$ . . .	·316	·564	·652	·551	·212	·371	·615
$r_{12·45}$ . . .	·367	·605	·496	·554	·367	·494	·667

TABLE III—*contd.*

Grain yield and thickness of earhead—(contd.)

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
$r_{12 \cdot 56}$ . . .	·517	·309	·614	·527	·611	·591	·706
$r_{12 \cdot 35}$ . . .	·455	·764	·815	·630	·282	·402	·653
$r_{12 \cdot 36}$ . . .	·395	·464	·787	·487	·216	·440	·627
$r_{12 \cdot 46}$ . . .	·278	·360	·429	·376	·341	·526	·647
Level of significance ($P = ·01$).	.165	·229	·164	·228	·162	·162	·162
$r_{12 \cdot 345}$ . . .	.319	·642	·659	·588	·228	·401	·646
$r_{12 \cdot 346}$ . . .	.215	·297	·340	·378	·147	·348	·621
$r_{12 \cdot 456}$ . . .	.284	·255	·426	·394	·354	·499	·673
$r_{12 \cdot 356}$ . . .	.298	·267	·784	·454	·220	·383	·659
Level of significance ($P = ·01$).	.166	·230	·165	·229	·162	·162	·162
$r_{12 \cdot 3456}$ . . .	.236	·331	·679	·370	·163	·380	·659
Level of significance ($P = ·01$).	.166	·231	·165	·229	·163	·163	·163

TABLE IV

Partial correlations

Grain yield and length of earhead

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
r_{13}	·748	·535	·701	·457	·828	·775	·847
Level of significance ($P = ·01$). .	·165	·227	·164	·226	·161	·161	·161
$r_{13·2}$ . . .	·437	·045	·726	—·123	·486	·490	·748
$r_{13·4}$ . . .	·206	—·006	·180	—·037	·332	·493	·498
$r_{13·5}$ . . .	·618	·333	·486	·281	·625	·564	·590
$r_{13·6}$ . . .	·747	·747	·711	·613	·851	·788	·855
Level of significance ($P = ·01$). .	·165	·228	·164	·227	·162	·162	·162
$r_{13·24}$ . . .	·130	—·058	—·052	—·208	·166	·307	·447
$r_{13·25}$ . . .	·404	.246	·689	—·010	·322	·368	·510
$r_{13·26}$ . . .	·530	·487	·785	·161	·554	—·127	·825

TABLE IV—*contd.*

Grain yield and length of earhead—(contd.)

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
$r_{13 \cdot 45}$ . . .	·191	·187	·181	·056	·330	·341	·410
$r_{13 \cdot 46}$ . . .	·303	·421	·325	·060	·393	·541	·512
$r_{13 \cdot 56}$ . . .	·622	·419	·555	·294	·671	·097	·620
Level of significance ($P = ·01$).	·165	·229	·164	·228	·162	·162	·162
$r_{13 \cdot 245}$ . . .	·106	·175	·663	—·100	·774	·137	·355
$r_{13 \cdot 246}$ . . .	·247	·371	·621	—·070	·253	·369	·469
$r_{13 \cdot 456}$ . . .	—·273	·382	·306	·080	·393	·387	·429
$r_{13 \cdot 256}$ . . .	·487	·381	·744	·091	·406	—·606	·670
Level of significance ($P = ·01$).	·166	·230	·165	·229	·162	·162	·162
$r_{13 \cdot 2456}$ . . .	·236	·360	·622	—·029	·243	·286	·515
Level of significance ($P = ·01$).	·166	·231	·165	·229	·163	·163	·163

The partial correlations obtained by eliminating the other factors though considerably reduced, except in *A. S.* 1063 (summer 1931) and *A. S.* 129 (main 1931), are nevertheless significant. That a partial correlation can be none too cautiously interpreted is well illustrated here. If L be the length of the earhead, and 2 t the thickness, the volume of an earhead, considering its shape, can be taken approximately as kt^2L. The grain yield is nearly proportional to this volume. If n be the packing in unit space and w the weight per grain, W, the total grain yield for the earhead is approximately kt^2Lnw. In this expression, n is the only undetermined factor and it may be supposed nearly constant. The low partial correlations are in contradiction of the obvious fact that, when for instance t and w are held constant, the correlation between W and L is perfect. This anomalous result is because the underlying assumption of linear regression is not fulfilled.

Diameter of peduncle and yield

The thickness or diameter of peduncle was measured with a Vernier callipers at a standard distance of 5 cm. below the base of panicle. The peduncle, which is virtually the end internode, does not show the amount of fluctuation in girth which the internodes lower to it, present.

TABLE V

Partial correlations

Grain yield and diameter of peduncle

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
r_{14}	·845	·739	·879	·569	·870	·855	·857
Level of significance (P=·01)	·165	·227	·164	·226	·161	·161	·161
$r_{14 \cdot 2}$ . . .	·617	·202	·611	·063	·664	·726	·776
$r_{14 \cdot 3}$ . . .	·617	·604	·746	·383	·557	·700	·541
$r_{14 \cdot 5}$ . . .	·760	·732	·742	·497	·659	·613	·577
$r_{14 \cdot 6}$ . . .	·823	·748	·828	·708	·885	·863	·870
Level of significance (P=·01)	·165	·228	·164	·227	·162	·162	·162
$r_{14 \cdot 23}$ . . .	·497	·205	·137	·181	·537	·661	·526
$r_{14 \cdot 25}$ . . .	·614	·500	·681	·247	·466	·524	·516
$r_{14 \cdot 26}$ . . .	·653	·400	·617	·354	·703	·743	·800

$r_{14 \cdot 35}$	·586	·704	·830	·430	·415	·511	·386
$r_{14 \cdot 36}$	·581	·427	·657	·452	·578	·451	·576
$r_{14 \cdot 56}$	·693	·244	·672	·499	·684	·674	·612
Level of significance ($P = ·01$)	·165	·229	·164	·228	·162	·162	·162
$r_{14 \cdot 235}$	·514	·475	·618	·266	·384	·483	·367
$r_{14 \cdot 236}$	·501	·225	·138	·341	·563	·692	·568
$r_{14 \cdot 356}$	·479	·161	·539	·428	·426	·051	·429
$r_{14 \cdot 256}$	·590	·213	·529	·282	·506	·609	·563
Level of significance ($P = ·01$)	·166	·230	·165	·229	·162	·162	·162
$r_{14 \cdot 2356}$	·441	·139	·088	·336	·403	—·023	·430
Level of significance ($P = ·01$)	·166	·231	·165	·229	·163	·163	·163

The diameter of peduncle is a very reliable index to yield [Rangaswami Ayyangar, 1932]. It is needless to add how easy and practical a guide this character is in the choice of seed material for yield.

The correlations of this character with the thickness and the length of the earhead are high. A thick peduncle is the mark of a large earhead. However, no *a priori* form of the dependence of yield on the diameter of the peduncle can be deduced; the correlation is indirectly through the length and thickness of the earhead. Table V shows the effect of eliminating the other factors step by step. The low values are not to be interpreted too literally for reasons already set forth.

Length of peduncle and yield

Varieties of sorghum vary in the length of their peduncles. The length of peduncle was taken to be the distance between the top-most node and the base of the earhead and measured in centimetres. It is seen from Table I that in four cases the correlations are below the level of significance, and that only in the case of *A. S.* 809 in the year 1932 the correlation value is a little high. The regression is however non-linear as is evident from the following analysis:—

A. S. 809. Summer 1932.

Analysis of variance

Variance	Degrees of freedom	Sum of squares	Mean square	$\frac{1}{2}$ Log$_e$ (Mean square)
Linear regression . . .	1	340·6	656·8	..
Deviations from regression .	4	72·0	18·0	1·445
Within arrays . . .	118	677·3	5·74	·874
Total .	123	1089·9	..	..

For $Z = ·571$, $P < ·05$

Dry straw weight and yield

After removing the head each stalk was folded up to a bundle about a foot and a half in length, and tied up with a cotton thread. These bundles were placed in wicker baskets and kept out for drying in the sun undisturbed. This minimised the loss due to shedding leaves. The weights of dry stalks were recorded after the bundles were thoroughly dry.

The correlations are higher in rain-fed than in irrigated varieties.

TABLE VI

Partial correlations

Grain yield and weight of dry straw

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
r_{15}	·588	·451	·702	·375	·763	·828	·835
Level of significance (P=·01) .	·165	·227	·164	·226	·161	·161	·161
$r_{15 \cdot 2}$ . . .	·225	—·274	·320	—·175	·559	·706	·778
$r_{15 \cdot 3}$ . . .	·286	·091	·489	·004	·436	·681	·549
$r_{15 \cdot 4}$ . . .	—·183	—·429	—·011	—·208	·170	·520	·489
$r_{15 \cdot 6}$ . . .	·617	·749	·655	·580	·781	·826	·838
Level of significance (P=·01) .	·165	·228	·164	·227	·162	·162	·162
$r_{15 \cdot 23}$ . . .	·133	·359	—·053	—·126	·439	·651	·584
$r_{15 \cdot 24}$ . . .	—·215	—·526	—·062	—·293	·194	·484	·526

$r_{15 \cdot 26}$ . . .	·355	·387	·375	·146	·595	·717	·788
$r_{15 \cdot 34}$ . . .	·050	—·461	—·020	—·212	·165	·472	·399
$r_{15 \cdot 36}$ . . .	·374	·428	·448	·171	·469	·741	·554
$r_{15 \cdot 46}$ . . .	—·045	·251	·061	—·034	·200	·541	·485
Level of significance ($P = \cdot 01$) .	·165	·229	·164	·228	·162	·162	·162
$r_{15 \cdot 234}$ . . .	—·067	—·558	—·125	—·233	·154	·463	·458
$r_{15 \cdot 236}$ . . .	·269	·218	—·014	·060	·470	·829	·523
$r_{15 \cdot 346}$ . . .	—·008	·164	—·045	—·064	·200	·500	·511
$r_{15 \cdot 246}$ . . .	—·093	·105	·013	—·116	·223	·517	·529
Level of significance ($P = \cdot 01$) .	·166	·230	·165	·229	·162	·162	·162
$r_{15 \cdot 2346}$ . . .	—·142	·046	—·055	—·098	·212	·475	·567
Level of significance ($P = \cdot 01$) .	·166	·231	·165	·229	·163	·163	·163

In the absence of more knowledge of the physiology of the sorghum plant the connection between straw weight and grain yield cannot be established by mere reasoning. Nevertheless it may be argued that the apparent relation between grain yield and straw can be traced to the common factor, the diameter of peduncle. A thick peduncle is the index of a thick stalk and generally of broad leaves, and therefore there is a high correlation between the diameter of the peduncle and the weight of straw ($r_{45} = .77$ to .83). The partial correlation coefficients, r_{15} .2346 in the irrigated varieties, are all not significant, and in the rain-fed varieties, they are considerably high, showing that in the varieties growing under dry conditions, straw is a factor to be reckoned with, independent of the other important factors.

Weight of 100 grains and yield

After the heads were threshed, 100 grains picked at random were weighed in grammes. This method was adopted to estimate the weight per grain, because of the impracticability of counting out all the grains in a head.

TABLE VII

Partial correlations

Grain yield and weight of 100 grains

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
r_{16}	·701	·805	·713	·731	·260	·394	·175
Level of significance ($P = ·01$) . .	·165	·227	·164	·226	·161	·161	·161
$r_{16 \cdot 2}$ . . .	·589	·726	·510	·733	·267	·348	·160
$r_{16 \cdot 3}$ . . .	·701	·884	·722	·795	·428	·446	·281
$r_{16 \cdot 4}$ . . .	·653	·811	·567	·810	·413	·443	·336
$r_{16 \cdot 5}$ . . .	·720	·898	·668	·800	·358	·380	·221
Level of significance ($P = ·01$) .	·165	·228	·164	·227	·162	·162	·162
$r_{16 \cdot 23}$ . . .	·648	·799	·633	·736	·397	·415	·283
$r_{16 \cdot 24}$ . . .	·630	·766	·518	·770	·401	·441	·342

TABLE VII—*contd.*

Grain yield and weight of 100 grains—(contd.)

r	Irrigated				Rain-fed Main 1931		
	A. S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
$r_{16 \cdot 25}$	·631	·745	·553	·730	·357	·385	·238
$r_{16 \cdot 34}$	·675	·898	·610	·811	·460	·499	·360
$r_{16 \cdot 35}$	·723	·906	·706	·802	·462	·456	·321
$r_{16 \cdot 45}$	·638	·780	·569	·801	·424	·450	·330
Level of significance ($P = \cdot 01$)	·165	·229	·164	·228	·162	·162	·162
$r_{16 \cdot 234}$	·651	·801	·633	·750	·438	·483	·372
$r_{16 \cdot 235}$	·672	·774	·651	·732	·432	·441	·339
$r_{16 \cdot 245}$	·609	·650	·468	·749	·414	·445	·349
$r_{16 \cdot 345}$	·529	·783	·673	·878	·471	·513	·354
Level of significance ($P = \cdot 01$)	·166	·230	·165	·229	·162	·162	·162
$r_{16 \cdot 2345}$	·494	·644	·719	·853	·429	·504	·376
Level of significance ($P = \cdot 01$)	·166	·231	·165	·229	·163	·163	·163

The irrigated varieties show a higher correlation than the rain-fed varieties. From the fact that in the former case the regressions are not far from linearity, the high correlations suggest a constancy of the number of grains per earhead; had the correlations been higher, this inference will be very conclusive. The partial correlations are high after eliminating all the other factors, showing that in selection, especially in the irrigated varieties, some consideration to the weight of 100 grains is worthwhile.

Duration and yield

Individuals in a variety having been noted to vary in the time of their flowering to within as much as a fortnight, an attempt was made to relate this difference to yield. Duration was calculated in days from sowing to the commencement of anthesis in the head concerned; the sowing date is more definite than the date of germination. This was recorded in two irrigated varieties for one season.

Within a variety, correlation between duration and yield of grain is negative and high (Table I), i.e., with the increase in duration there is a corresponding decrease in yield. This points to the desirability of compressing if possible the range of flowering period. Moreover, in selection work, there is no need to wait for the late plants; the earlier ones are likely to be better yielders than those delaying.

Duration is very important in an irrigated variety and it is not so great a limitation with a rain-fed variety. For this reason, duration was not considered in the rain-fed varieties.

Weight of earhead, fresh and dry, and yield

The weight of the earhead includes the weight of the standard 5 cm. stalk with which it was cut. The weight of the fresh head was recorded soon after cutting and the heads were kept for drying and their dry weights recorded after they were thoroughly dried for a number of days, special care being taken to avoid the shedding of grain.

Table I shows that the correlations between grain yield and the weight of earhead, fresh and dry, give the highest values. The differences in values between fresh and dry weights are negligible. It would thus be obvious that the weight of the earhead is a sufficiently reliable index of grain weight.

Table VIII presents the multiple correlation coefficients of grain yield with the other characters, beginning with $r_{1.4}$ which is of primary importance because of its high value, and the practicability of considering the diameter of peduncle in actual selection work.

TABLE VIII

Multiple correlations

R	Irrigated				Rain-fed Main 1931		
	A.S. 809		*A. S.* 1063				
	Summer 1931	Summer 1932	Summer 1931	Summer 1932	*A. S.* 60	*A. S.* 1098	*A. S.* 129
r_{14}	·845	·739	·879	·569	·870	·855	·857
Level of significance ($P = ·01$) .	·165	·227	·164	·226	·161	·161	·161
$R_{1·24}$ . . .	·866	·824	·910	·714	·887	·892	·916
Level of significance ($P = ·01$) .	·193	·266	·192	·265	·190	·190	·190
$R_{1·234}$ . . .	·868	·824	·934	·722	·890	·904	·934
Level of significance ($P = ·01$) .	·214	·294	·213	·293	·210	·210	·210
$R_{1·2345}$ . . .	·874	·896	·935	·746	·890	·924	·947
$R_{1·2346}$ . . .	·900	·943	·986	·948	·912	·937	·943
Level of significance ($P = ·01$) .	·231	·317	·230	·316	·227	·227	·227

The addition of variates increases R, in some cases significantly and in some cases not. However, throughout ${}^{R}1{\cdot}2345$ and ${}^{R}1{\cdot}2346$ are significantly above ${}^{r}14$. The tests of s gnificance presented in Table IX were carried out by considering P corresponding to ${}^{z}1{\cdot}234p - 14 = \frac{1}{2}\log_e \frac{({}^{R^2}1{\cdot}234p - {}^{r^2}14)\ (N-5}{3(1-{}^{R^2}1{\cdot}234p)}$ ($p=5, 6$) for degrees of freedom $n_1=3$ and $n_2=N-5$ where N is the size of the samples [Tippett, 1931]. ${}^{R}1{\cdot}2346$ tends to be higher than ${}^{R}1{\cdot}2345$, thus indicating that given ${}^{R}1{\cdot}234$, the weight of 100 grains is more informative than the weight of straw which is less easy to handle. It may be said, that the additional factors, *viz.*, the length and thickness of earhead, and the weight of 100 grains, help to predict grain yeild with a greater precision than is possible with a knowledge of the diameter of peduncle only.

TABLE IX

Tests of significance

Strain	Season	$z_{1 \cdot 2345—14}$	Degrees of freedom		P	$z_{1 \cdot 2346—14}$	Degrees of freedom		P
			n_1	n_2			n	n_2	
A. S. 809 . .	Summer 1931 .	1·403	3	235	$<$·01	1·730	3	235	$<$·01
A. S. 809 . .	Summer 1932 .	1·972	3	119	$<$·01	2·406	3	119	$<$·01
A. S. 1063. .	Summer 1931 .	2·080	3	238	$<$·01	3·172	3	238	$<$·01
A. S. 1063. .	Summer 1932 .	1·522	3	120	$<$·01	2·713	3	120	$<$·01
A. S. 60 . .	Main 1931 . .	1·313	3	245	$<$·01	1·796	3	245	$<$·01
A. S. 1098. .	Main 1931 . .	2·114	3	245	$<$·01	2·294	3	245	$<$·01
A. S. 129. .	Main 1931 . .	2·109	3	245	$<$·01	2·369	3	245	$<$·01

Review of previous work

The above correlation studies made through three seasons on the Coimbatore varieties, both dry and irrigated, lead us to a review of previous work in this field. So far, correlation studies in Indian sorghums have been reported only in a few Bombay varieties. Patel [1923] tentatively characterised an ideal *jowar* plant as having a large number of nodes and thus a corresponding increase in the number and total leaf surface per plant. The plant must be tall with the top five internodes short, and having a compact earhead and increased number of seeds per spike. Each of these characters bears a high degree of correlation to grain yield. Kottur and Chavan [1927, 1928] determined the correlations between yield and seven other characters and found that the correlations are positive and reliable in five, *viz.*, height of plant, number of leaves, length of rachis, breadth and weight of the ripe ear, and small and unreliable in two cases, *viz.*, length of peduncle and size of the grain. Patel and Patel [1927-28] working on Surat *jowars* arrived at the same conclusion as above.

In the Texas Stations Report for 1928, the following observations occur:— "Correlation coefficients determined in Blackhull Kafir indicated that length of head, length of seed branches, diameter of plants, and weight of green forage are the characters contributing significantly towards production of grain yield." Harper and others [1931] report from Oklahoma that "inheritance studies with sorghum indicated that length of seed branches and the circumference of heads might be associated closely with high yields, while the length of the stalk had little to do with the grain yield." Unlike the above workers Martin [1928] who has reported on plant characters and acreage yields of grain sorghums concludes among other things that "the yields of fields of grain sorghums are more closely correlated with the number of heads per acre than with the size of head or weight of grain per head. The correlation between the number of heads per acre and both the weight per bushel of grain and the average size of heads is either negative or not significant in the three varieties studied. The height of stalks within a given variety is highly correlated with grain yields. The yields of grain and stover in Kafir are closely correlated. The size or plumpness of seeds of a given variety is correlated with weight per bushel."

In our studies, we determined the relationship of nine characters with grain yield. Our results in general agree with those of the Bombay workers for all the characters common to our studies. A feature of our work is an attempt at gauging the comparative behaviour of rain-fed and irrigated varieties grown under different agronomic conditions. The greater elaboration with which these relationships were pursued have now enabled us, armed with that knowledge, to safely dispense with certain of them and seek in the pursuit of a few significant plant characters, valuable aids to yield.

Summary

The correlation between grain yield per plant and eight other plant characters have been determined in two irrigated and three rain-fed Coimbatore varieties of sorghum.

The diameter of peduncle, weight, length and thickness of earhead and straw weight have given high positive correlation values. These characters can be used as reliable indices in selecting for high yield.

The weight of 100 grains has given high correlation values in the irrigated varieties while in the three dry varieties it was low.

The length of peduncle is either not correlated or is negatively correlated with yield.

In the two irrigated varieties studied the duration was found to be negatively correlated with yield.

Partial and multiple correlations were also calculated. The total grain yield of a plant can be predicted very closely, when the diameter of peduncle, length and thickness of earhead, and the weight of 100 grains are all known.

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ABSTRACT

Heterosis as a factor in the increase of tobacco yield. 633·71 :575·125

E. A. Osadchuk. (*Junzo-bereznoc Otdelenie V. I. R'a Gosudarstennyi Nikitskii Botaniceskii sad, Nikita Botanic Garden, 1934: Bull. No. 12, pp. 40*) (*Translation of full summary of the paper, issued by the Imperial Bureau of Plant Genetics, School of Agriculture, Cambridge, England*).

The F_1 hybrids of a number of parental combinations were compared with the parents in two successive years, 1929 and 1930 (Table 3, pages 102 and 103).

Not by any means all combinations shewed heterosis; hybrids of varieties genetically closely allied were invariably intermediate, those from unrelated varieties generally shewed at least an increase in height.

Descriptions are given of the varieties used as parents, followed by tables shewing the yields of the parent varieties and F_1 hybrids. The best combination, Nikitskii Dubec 44 × Sumatra Petioled, gave an increase of 35·1 per cent over the better of the two parents, the next best American 572 × Sumatra Petioled, 26·9 per cent; each of these crosses involved (Table 7, page 104) a Sumatra tobacco and a local strain. Two out of the seven combinations gave negative results, *i.e.*, reduced yields.

The same hybrids were grown on a second plot where the soil fertility was much lower and none of them shewed any marked yield increases: the best combination in the first plot here gave a yield intermediate between the parents and the second best shewed a decrease of 11·7 per cent in place of a 26·9 per cent increase. It is evident from this that it is necessary to test the hybrids under a variety of conditions before drawing conclusions regarding heterosis and to do this a system of tests at a series of different centres, analogous to the State variety testing systems, is regarded as necessary.

Similar studies were made on the number of leaves fit for picking. This varied very little in any one variety and the hybrids were mostly intermediate. The two most highly yielding hybrids referred to above had a greater number of leaves than the parents in both plots. The middle leaf of (Table 8, page 105) 50 plants of each type was measured, the length, breadth, area, form (length: breadth) and thickness of the leaves being determined (Table 9, page 106). The leaf area exceeded that of the parents in several of the hybrids, in one case by 17 4 per cent. American 572 × Sumatra Petioled had actually smaller leaves though their number was greater. Only one combination was intermediate in leaf area.

The yields estimated by multiplying the number, size and thickness of the leaves in most cases corresponded fairly closely to the actual yields and this is suggested as a useful method of making yield comparisons, especially in experiments where only one variety is in use.

The yields of first, second and third quality product and degustation tests shewed that though the differences between the varieties were not great, many of the hybrids excelled their parents and the superiority was most pronounced in the combinations shewing most increase in yield. In view of this fact and the ease of effecting the crosses the use of first generation hybrids on a much greater scale is recommended. It is calculated that one trained worker can produce enough seed to sow one ha. in two working days.

TABLE 3

Results of investigation of the F_1 hybrids in comparison with their parent forms

Parent forms Combination	Percentage increase in 1929				Percentage increase in 1930			
	Height		Number of leaves		Height		Number of leaves	
	In comparison with mean yield of parents	In comparison with the higher yielding parent	In comparison with mean yield of parents	In comparison with the higher yielding parent	In comparison with mean yield of parents	In comparison with the higher yielding parent	In comparison with mean yield of parents	In comparison with the higher yielding parent
1. American 572 × Sumatra Petioled	+27	+20	−3	−15	+6	+6	+8	0
2. Dubec Autskii 576 × Havana	+15	+15	+27	+11	+14	+4	+11	+3
3. Dubec Nikitskii 44 × Sumatra Petioled	+16	+6	+9	−5	+7	—4	0	—6
4. Dubec Nikitskii 44 × American 572	+34	+6	+3	−5	+22	+9	+4	−5
5. Deli × Havana	+13	+4	+4	+2	+30	+30	+12	+10
6. Havana × Ameraicn 572	+16	+3	+10	−3	+17	+12	+12	+4
7. Aya Suluk × Havana	+12	+1	+21	+3	+2	−8	−9	−27
8. Dubec Derekoiskii × Havana	+13	+8	+3	−4	+11	+4	0	−19

Parent forms Combination	Percentage increase in 1929				Percentage increase in 1930			
	Height		Number of leaves		Height		Number of leaves	
	In comparison with mean yield of parents	In comparison with the higher yielding parent	In comparison with mean yield of parents	In comparison with the higher yielding parent	In comparison with mean yield of parents	In comparison with the higher yielding parent	In comparison with mean yield of parents	In comparison with the higher yielding parent
9. Sumatra Petioled × Varatik	+13	0	+4	−4	0	−15	−8	−18
10. Persicăn × Dubec Nikitskii 44	+28	−1	+8	−9	+6	−7	+3	−14
11. Ksanti × Dubec Derekoiskii	+6	−3	−3	−11	..	..	..	..
12. Sandre × Dubec Nikitskii 44	+1	−4	+7	−6	−12	−10	+6	−13
13. Kizildza Mursal × Dubec Nikitskii 44	−3	−12	−11	−26	+10	−3	0	−7
14. Havana × Dubec Gurzufskii 282	−1	−14	0	−20	+7	−17	−7	−20
15. Havana × Dubec Nikitskii 44	+15	−9	+7	−11	+18	+3	−8	−12
16. Varatik × Bektemiz	+2	−20	−11	−27	−20	−37	−13	−43
17. Ksanti × Havana	..	−3	..	..	+22	+12	+15	+11
18. American 572 × Aya Suluk	..	..	..	..	+11	+7	−7	−10
19. Dubec Gaspra × American 572	..	..	..	..	+5	5	−3	−9
20. Dubec Gurzufskii × Sansun 84	..	..	..	..	0	−10	−10	−23
21. Havana × Varatik	..	..	..	..	0	−10	−14	−14

TABLE 7

Yield of air-dried leaves in centners per hectare

	Name of varieties and combinations	$M \pm m$	P	Percentage increase	
				Over the mean	Over the higher parent
	Plot 1				
	Deli	20·3±0·52	2·6	..	..
F_1	Deli × Havana	18·8±0·64	3·4	−1·6	−7·4
	Havana	12·2±0·69	5·7	..	..
F_1	Dubec Autskii 576 × Havana	14·5±0·67	4·6	+1·0	−2·0
	Dubec Autskii 576	14·8±0·49	3·3	..	..
	Dubec Nikitskii 44	14·5±0·41	2·8	..	..
F_1	Havana × Dubec Nikitskii 44	16·4±0·44	2·7	+23·3	+13·1
F_1	Dubec Nikitskii 44 × American 572	19·9±0·66	3·3	+24·5	+16·4
F_1	Havana × American 572	19·6±0·55	2·8	+34·2	+14·6
	American 572	17·1±0·85	2·1	..	..
F_1	American 572 × Sumatra Petioled	21·7±0·49	2·3	+33·1	+26·9
F_1	Dubec Nikitskii 44 × Sumatra Petioled	21·1±0·93	4·4	+40·7	+36·1
	Sumatra Petioled	15·5±0·55	3·6	..	..
	Plot 2				
	Deli	14·8±0·42	2·8	..	..
F_1	Deli × Havana	13·8±0·94	6·8	+2·5	−0·66
	Havana	12·2±0·50	4·1	..	..
F_1	Dubec Autskii 576 × Havana	13·6±0·69	5·1	+4·7	+2·3
	Dubec Autskii 576	13·3±0·77	5·8	..	..
	Dubec Nikitskii 44	15·4±0·42	2·7	..	..
F_1	Havana × Dubec Nikitskii 44	12·4±0·72	5·8	+1·0	−1·9
F_1	Dubec Nikitskii 44 × American 572	17·3±0·99	5·7	+1·1	+0·7
F_1	Havana × American 572	17·8±0·75	4·2	+26·2	+1·1
	American 572	16·1±0·82	5·4	..	..
F_1	American 572 × Sumatra Petioled	14·2±0·74	5·2	−9·5	−11·7
F_1	Dubec Nikitskii 44 × Sumatra Petioled	15·6±0·71	4·6	+0·13	+0·13
	Sumatra Petioled	15·4±0·36	2·3	..	..

TABLE 8

Mean number of leaves picked per plant

Varieties and combinations	Plot 1			Plot 2		
	Total number of leaves	Number of leaves picked per plant	Mean number of leaves per plant	Total number of leaves	Number of leaves picked per plant	Mean number of leaves per plant
Deli . . .	13725	909	15·1	13574	911	14·9
F_1 Deli × Havana .	13379	904	14·8	13890	926	15·0
Havana . .	11986	908	13·3	11164	886	12·6
F_1 Dubec Autskii 576 × Havana	19367	905	21·4	18444	891	20·6
Dubec Autskii 576	20498	907	22·6	18339	899	20·4
Dubec Nikitskii 44	22213	885	23·1	20114	890	22·6
F_1 Havana × Dubec Nikitskii 44	21382	906	23·6	20384	910	22·4
F_1 Dubec Nikitskii 44 × American 572	20996	915	23·2	19276	905	21·3
F_1 Havana × American 572	15305	911	16·8	14667	906	16·2
American 572 .	14579	889	16·4	12879	810	15·4
F_1 American 572 × Sumatra Petioled	20648	840	23·2	18931	919	20·6
F_1 Dubec Nikitskii 44 × Sumatra Petioled	23135	883	26·2	23025	921	25·0
Sumatra Petioled .	15901	910	17·4	15205	916	16·6

TABLE 9

Mean data on dimensions of the middle leaf in the forms investigated

1	Name of varieties and combinations 2	Area in sq. cm. 3	Ratio of length to breadth 4	Mean weight of unit area of leaf 5	Yield calculated from size and thickness of leaf 6	Percentage increase on higher yielding forms 7	Percentage increase on higher yielding forms by calculation 8	Difference between columns 7 and 8 9
	Deli	197	2·27	196	11837	..	..	..
F_1	Deli × Havana	212	2·43	316	12594	+6	−7·4	1·4
	Havana	132	3·28	320	5153	..	..	..
F_1	Dubec Autskii 576 × Havana	148	2·37	391	6034	−7	−2·0	5·0
	Dubec Autskii 576	123	2·19	233	6478	..	..	..
	Dubec Nikitskii 44	114	2·48	246	4066	..	..	..
F_1	Havana × Dubec Nikitskii 44	165	2·46	315	8523	+11	+13·1	2·1
F_1	Dubec Nikitskii 44 × American 572	163	2·12	254	7162	+13	+16·4	3·4
F_1	Havana × American 572	156	2·10	233	7124	+25	+14·6	10·4
	American 572	149	2·32	223	5682	..	..	..
F_1	American 572 × Sumatra Petioled	142	2·35	239	7364	+29	+26·9	3·9
F_1	Dubec Nikitskii 44 × Sumatra Petioled	166	2·14	260	11307	+40	+36·1	3·9
	Sumatra Petioled	184	2·28	248	7980	..	..	..

NOTES

INDIGENOUS DRUGS OF INDIA

In the course of the discussion on two applications for research grants for investigations on indigenous drugs, it was brought to notice that information was not conveniently available on Indian drug-producing plants. Col R. N. Chopra, I.M.S., Professor of Pharmacology, School of Tropical Medicine, Calcutta, has kindly furnished the following lists of:—

A. Important indigenous drugs for which there is a good market.

B. Pharmacopoeial drugs or their substitutes, which grow in India.

Further details will be found in Col. Chopra's book "The indigenous drugs of India".

LIST OF IMPORTANT INDIGENOUS DRUGS GROWING OR CULTIVATED IN INDIA FOR WHICH THERE IS A GOOD MARKET

1. *Abroma augusta*—throughout India.
2. *Abrus precatorius*—throughout India.
3. *Acalypha indica*—throughout India.
4. *Achillea millefolium*—Western Himalayas, cultivated in gardens.
5. *Achyranthes aspera*—throughout India.
6. *Adhatoda vasica*—throughout India.
7. *Aegle marmelos*—throughout India.
8. *Aesculus hippocastanum*—in the Punjab.
9. *Agave americana*—throughout India.
10. *Alangium lamarckii*—throughout India.
11. *Alpinia galanga*—E. Bengal and South India.
12. *Alstonia scholaris*—throughout India.
13. *Amomum subulatum*—throughout India.
14. *Anacyclus pyrethrum*—throughout India.
15. *Anamirta cocculus*—Concan, Malabar, E. Bengal and Assam.
16. *Andrachne cordifolia*—Punjab.
17. *Andrographis paniculata*—throughout India.
18. *Andropogan iwarancusa*—cultivated throughout India.
19. *Anona squamosa*—throughout India.
20. *Anthemis nobilis*—Northern India.
21. *Antiaris toxicaria*—Burma.
22. *Argemone mexicana*—throughout India.

23. *Aristolochia bracteata.*
24. *Bassia latifolia*—throughout India.
25. *Berberis aristata, asiatica*—Himalayas, Nilgiris and Ceylon.
26. *Blumea lacera*—throughout India.
27. *Boerhaavia diffusa*—throughout India.
28. *Butea frondosa*—throughout India.
29. *Caesalpinia bonducella*—throughout India.
30. *Calotropis gigantea*—throughout India.
31. *Cedrus libani*—throughout India.
32. *Cuminum cyminum*—throughout India.
33. *Datura stramonium*—Punjab.
34. *Euonymus crenulatus*—Temperate and Western Himalayas.
35. *Euphorbia pillulifera*—throughout India.
36. *Fumaria officinalis*—Chittoor and Punjab.
37. *Gaultheria fragrantissima*—Nilgiris.
38. *Gossypium herbaceum*—throughout India.
39. *Hedyotis auricularia.*
40. *Helleborus niger*—throughout India.
41. *Holarrhena antidysenterica*—throughout India.
42. *Hygrophila spinosa*—throughout India.
43. *Ipomaea hederaceae*—throughout India.
44. *Ipomaea turpethum*—throughout India.
45. *Mallotus philippinensis*—throughout India.
46. *Melia azadirachta*—throughout India.
47. *Ocimum basilicum*—throughout India (*O. gratissinum*).
48. *Oldenlandia biflora*—throughout India.
49. *Oroxylum indicum*—throughout India.
50. *Peganum harmala*—N.-W. India, Sind, Punjab, Kashmir, Agra and Western Deccan.
51. *Petroselinum sativum, P. graveolensis.*
52. *Picrasma quassioides*—subtropical Himalayas and Kashmir.
53. *Piper cubeba, P. betle*—throughout India.
54. *Plumbago rosea*—throughout India.
55. *Pongamia glabra*—throughout India.
56. *Psoralia corylifolia*—throughout India.
57. *Quercus infectoria*—scattered throughout India.

58. *Rauwolfia serpentina*—tropical Himalayas and at the foot of the hills from Moradabad to Sikkim, Assam, Pegu, Travancore and Ceylon.
59. *Saraca indica*—throughout India.
60. *Scopolia lurida*—Central Himalayas, Nepal, Sikkim.
61. *Semicarpus anacardium*—throughout India.
62. *Sida cordifolia*—throughout India.
63. *Solanum dulcamara*, *S. indicum*—throughout India.
64. *Swertia chirata*—from Kashmir to Bhutan and Khasia range, at 4,000—10,000 feet.
65. *Symplocos racemosa*—throughout India.
66. *Terminalia arjuna*—throughout India.
67. *Tinospora cordifolia*—throughout India.
68. *Toddalia aculeata*—Nilgiris and from Kumaun to Bhutan, upto 5,000 feet.
69. *Tribulus terrestris*—throughout India and Ceylon.
70. *Tylophora asthmatica*—Madras.
71. *Vernonia anthelmintica*—throughout India.
72. *Viola odorata*—Kashmir, Northern India.
73. *Withania somnifera*—dry subtropical India, west coast.

LIST OF PHARMACOPOEIAL DRUGS OR THEIR SUBSTITUTES GROWING IN INDIA

1. *Acacia arabica*—Indian substitute *Acacia indica*—throughout India.
2. *Aconitum napellus*, grows abundantly. *A chasmanthum Stapf.*—In Kashmir, Bhutan and Tibet.
3. *Aloe chinensis* (*A. perry* and other species, specially thyrox)—Southern India. Substitute *A. indica*.
4. *Arachis hypogoea*—throughout India.
5. *Artemisia maritima*—from Kumaun to Kashmir at a height of 4,000 to 12,000 feet. More abundantly in Baluchistan, Chitral, Afghanistan and in the Himalayas.
6. *Astragalus fucumifr*—imported from Persia. Indian substitute *A. heratensis*, *A. strobiliferus*.
7. *Atropa belladonna*—Western Himalayas.
8. *Balsamodendron myrrha* and other species—Central Provinces, Cutch and Sind.
9. *Brassic nigra*, *B. campestris* and *B. juncea*—cultivated universally.

10. *Camellia Thea* and *Coffea arabica*—grow in Darjeeling, Assam, Sylhet, Ceylon and the forests of Southern India.
11. *Cannabis sativa* (or *indica*)—throughout India.
12. *Capsicum*—throughout India.
13. *Carum copticum*—throughout India.
14. *Carum carui*—throughout India.
15. *Cassia fistula*—throughout India.
16. *Cassia augustifolia* and *acutifolia*—throughout India.
17. *Chenopodium ambrosoides*, var. *anthelminticum*—Baluchistan.
18. *Cinchona succirubra* and other varieties—Sikkim, Nilgiri hills, Mungpoo and Ceylon.
19. *Cinnamomum camphora*—bulk imported in India.
20. *Citrus aurantium*—throughout India.
21. *Citrullus colocynthis*—North-West, Central and South India.
22. *Colchicum autumnale* and *luteum*—Kashmir and common in Badghis and Khorasan.
23. *Convolvulus scammonia*.
24. *Coriandrum sativum*—throughout India.
25. *Digitalis purpurea*—cultivated in Kashmir.
26. *Dorema ammoniacum* and other species—Baluchistan.
27. *Elettaria cardamomum*—Western and Southern India.
28. *Eugenia caryophylata*—Malabar coast and Ceylon.
29. *Ephedra vulgaris* and allied species—dry stony hills of north-west Afghanistan, Baluchistan, intermediate Himalayas, Kumaon.
30. *Eucalyptus globulus*—Himalayas.
31. *Ferula foetida*, Indian substitute *F. narthex*—Afghanistan.
32. *Foeniculum vulgare*—cultivated throughout India.
33. *Gentiana lutea*, Indian substitute *G. kurroa*—Kashmir, north-west Himalayas.
34. *Glycyrrhiza glabra*—introduced into Punjab and Sind.
35. *Hyoscyamus niger* and *muticus*—Kashmir and north-west Himalayas.
36. *Juniper communis*, *macropoda*—western Himalayas.
37. *Linum usitatissimum*—cultivated throughout India.
38. *Lobelia inflata*, Indian substitute *nicotianifolia*—Bombay to Travancore and Ceylon.
39. *Mentha arvensis* and *piperita*—Western Himalayas, also cultivated in gardens throughout India.

40. *Myristica fragrans*—cultivated in South India.
41. *Papaver somniferum*—Bihar, U. P. and Punjab.
42. *Peucedanum graveolens*—throughout India.
43. *Picroena excelsa*—Kashmir and north-west Himalayas.
44. *Picrorrhiza kurrooa*—Alpine Himalayas from Kashmir to Sikkim.
45. *Pipenella anisum*—cultivated throughout India.
46. *Pinus longifolia*—Himalayas.
47. *Podophyllum emodi*—N.-W. Himalayas.
48. *Polygala senega*, Indian substitute *P. chinensis, crotalariodes*—throughout India.
49. *Psychotria ipecacuanha*—cultivated in Nilgiris, Mungpoo, Burma.
50. *Ricinus communis*—cultivated throughout India.
51. *Rheum emodi*—Himalayas.
52. *Santalum album*—South India.
53. *Strophanthus.*
54. *Strychnos nux vomica*—specially South India, and Cuttack in B. & O.
55. *Taraktogenos kurzii* and *Hydnocarpus wightiana*—Western Peninsula, South Concan to Travancore.
56. *Thymus vulgaris* (*Corum copticum*)—Indore and Nidam's Domisions.
57. *Urginea indica*—throughout India.
58. *Valeriana wallichii* and *officinale*—N.-W. Himalayas.
59. *Zingiber officinale*—cultivated throughout India.

THE MAYNARD-GANGA RAM PRIZE

In 1925 the late Sir Ganga Ram, Kt., C.I.E., M.V.O., R.B., Lahore, with that generosity for which he was so well known, handed over to the Punjab Government a sum of Rs. 25,000 for the endowment of prize of the value of Rs. 3,000 to be called the Maynard-Ganga Ram Prize and to be awarded every three years, for a discovery, or an invention, or a new practical method which will tend to increase agricultural production in the Punjab on a paying basis. The competition is open to all throughout the world. Government servants are also eligible to compete for it.

Entries for the next award were invited by the 31st December, 1933. None of the entries was considered to be of sufficient merit and it has been decided by the Managing Committee of the prize that the award should be postponed for another year and that further entries should reach the Director of Agriculture, Punjab, Lahore, on or before the 31st December, 1935.

Applications are invited for the Maynard-Ganga Ram Prize of the value of Rs. 3,000 which will be awarded for a discovery, or an invention, or a new practical method tending to increase agricultural production in the Punjab on a paying basis. The prize is open to all, irrespective of caste, creed or nationality and Government servants are also eligible for it. Essays and theses are not eligible for competition and applicants should prove that some part of their discovery, invention, etc., is the result of work done after the prize was founded in 1925. The Managing Committee reserves to itself the right of withholding or postponing the prize, if no satisfactory achievement is reported to it. All entries in competition for the next award should reach the Director of Agriculture, Punjab, Lahore, on or before the 31st December, 1935.

THE KATAMORPHISM OF IGNEOUS ROCKS UNDER HUMID TROPICAL CONDITIONS

BY

THE LATE PROFESSOR SIR JOHN BURCHMORE HARRISON, KT., C.M.G., M.A., F.I.C., F.C.S., F.G.S., F.G.S.A., ETC.,

Late Director of Science and Agriculture, Government Analyst and Geologist, British Guiana

(Published by The Imperial Bureau of Soil Science, Rothamsted Experimental Station, Harpenden—Price 5*s*.)

Sir John Burchmore Harrison, late Director of the Department of Science and Agriculture in British Guinea, died in 1926, leaving behind him the unrevised manuscript of what was perhaps his greatest work. Throughout the 37 years he spent in British Guiana he interested himself in the study of the processes of tropical weathering to which he directed his great powers of observation and analysis. He incorporated the results and conclusion of his life's work on the subject in a manuscript which was discovered by Mr. R. R. Follett Smith of the British Guiana Department of Agriculture in 1930, and forwarded to the Soil Bureau by Professor F. Hardy, of the Imperial College of Tropical Agriculture. The Bureau does not undertake the publication of original research work, but in this instance it has been privileged to do so on behalf of Demerara Proprietors, Ltd., and the British Association for the Advancement of Science, who have generously contributed the necessary funds. The laborious and difficult task of revising the manuscript after the death of its author has been carried out by Professor Hardy. The scientific world owes a debt of gratitude to the contributing bodies and to Professor Hardy for securing the publication of a work of unique value which could never have been printed without their assistance. It is doubtful whether such a detailed and far-reaching study of tropical soil-forming processes, in particular of lateritisation, has ever been made before, and (under present-day conditions) it is most unlikely that the opportunity and ability to repeat Harrison's work will again occur together. It may truthfully be said that it scarcely needs repetition, but it will serve as a sure foundation to further researches on a problem of increasing scientific and economic importance. The monograph should become a standard work of reference to all students of tropical geology and soils.

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Crown quarto; 80 pp., with a foreword by Sir E. J. Russel and a preface by Professor Hardy. Stiff paper cover, Price 5 Shillings, post free.

APPENDIX

INSTRUCTIONS TO AUTHORS OF PUBLICATIONS OF THE IMPERIAL COUNCIL OF AGRICULTURAL RESEARCH*

1. All manuscripts should be clean, clear and carefully revised. Only one side of the paper should be used, and as far as practicable the original type-written copy and not a carbon copy should be sent. Capitals should be sparingly used, and all the necessary punctuations should be done in the MS. and not left for introduction in proofs.

2. The title of a paper should not be lengthy.

3. It is desirable that the MS. should have suitable heads and sub-heads. In numbering the principal divisions of a paper roman numerals should be used. The use of arabic figures and (*a*), (*b*), (*c*), etc., is generally reserved for numbering the sub-divisions coming under each head.

4. Articles submitted for publication either in the *Indian Journal of Agricultural Science* or in the *Indian Journal of Veterinary Science and Animal Husbandry* should be accompanied by abstracts for publication in *Agriculture and Live-stock in India*. Abstracts should be concise, but should be long enough to explain the matter dealt with; ordinarily no abstract should exceed 200 words.

5. When a word or line is intended to be printed in *italics* it should be underlined with a single line, in SM. CAP. with two lines, in CAPITALS with three lines, and when in **Antique** (heavy type) with a wavy line (~~~~~~~~).

6. In descriptive matter, numbers under 100 and all numbers occurring at the beginning of a sentence should be in words.

7. Local names for crops, technical operations, etc., should be defined where they first occur in the text, *e.g.*, *rabi* (spring crop). The use of local weights and measures should be avoided as far as possible. Vernacular names, such as *jowar*, *bajri*, should be in italics without a capital letter, and each such name where it first occurs should be followed by its scientific equivalent in brackets, *e.g.*, *jowar* (*Andropogon Sorghum*). It is usual to write the initial letters of varietal names in capitals, *e.g.*, Striped Mauritius, Dharwar-American cotton and Broach cotton.

8. Botanical and zoological names are printed in italics and should be underlined in the MS., *e.g.*, *Triticum vulgare* L.; *Diplodia Corchori* Syd.; *Pyrilla aberrans* Kirby. The International Rules of Botanical Nomenclature and the International Rules of Zoological Nomenclature should be followed. The names of chemical substances should not be written with a capital letter; they are printed in roman type (*e.g.*, calcium carbonate, prussic acid).

* Spare copies of these Instructions can be had on application to the Secretary, Imperial Council of Agricultural Research (Publication Section), New Delhi.

9. The following and similar abbreviations may be used freely:—*viz.*, *e.g.*, *i.e.*, mm. (millimetre), cm. (centimetre), grm. (gramme), mg. (milligramme), c.c. (cubic centimetre), sp. gr. (specific gravity), lb. (pound), cwt. (hundredweight), in. (inch), ft. (foot), oz. (ounce), md. (maund), sr. (seer), ch. (chattack). Other abbreviations should be used sparingly, if at all.

10. References to plates should be given within brackets, without prefixing the word "see" or "cf.", in the MS. itself, and should not be left over for introduction in proofs. For example, "The parasite (Pl. X, fig. 4) was present late in 1906".

11. The word "Table" is preferable to "Statement", and tables should be numbered consecutively in roman figures. Each table should have an explanation as a sub-head. It is more convenient for reference if tables can be printed horizontally; for this purpose they should not exceed in width the printing measure of the page (5 in.). *Example*—

TABLE IV

Results of water-saving experiments on wheat (Pusa 12) at Gungapur, Haripur and Sargodha, 1916-17

Station	No. of irrigations including the preliminary watering	Yield per acre in maunds and seers		Average yield per acre	
		Grain	Straw	Grain	Straw
		Mds. Srs.	Mds. Srs.	Mds. Srs.	Mds. Srs.
Gungapur	One	12 19½	20 10	9 34	21 17
Haripur	,,	8 31	19 14		
Sargodha	,,	8 12½	25 27½		

12. References to literature, arranged alphabetically according to author's names, should be placed at the end of the article, the various references to each author being arranged chronologically. Each reference should contain the name of the author (with initials), the year of publication, the abbreviated title of the publication, volume and page. In the text the reference should be indicated by the author's name followed by the year of publication enclosed in brackets; when the author's name occurs in the text, the year of publication only need be given in brackets. If reference is made to several articles published by one author in a single year, these should be numbered in sequence and

the number quoted after the year both in the text and in the collected references. This system of referencing is the same as is used in the *Biochemical Journal* with slight modification and will be clear from the following illustration:—

The work of Osborne and Mendel [1919, 1, 2] and Steenbock and Boutwell [1919] had indicated an association of the fat-soluble vitamin with the green parts of plants. This view was examined by Coward and Drummond [1921] who reported that vitamin A was not synthesised by etiolated shoots but that green leaves were active in its formation. Another worker [Wilson, 1922], on the other hand, found that etiolated shoots if given in sufficient quantity could supply the fat-soluble vitamin and that this factor was therefore formed in the absence of light.

REFERENCES

Coward, K. H. and Drummond, J. C. (1921). *Biochem. J.* **15**, 530.
Osborne, T. B. and Mendel, L. B. (1919, 1). *J. Biol. Chem.* **37**, 187.
——————————(1919, 2). *J. Biol. Chem.* **41**, 549.
Steenbock, H. and Boutwell, R. (1919). *J. Biol. Chem.* **41**, 149.
Wilson, J. (1922). *J. Biol. Chem.* **51**, 455.

Abbreviations, as far as possible, should follow the system adopted in "A World List of Scientific Periodicals" published by the Oxford University Press.

13. Papers should be complete when submitted for publication. As alterations and additions at the proof stage cause both additional expense and delay, they should be resorted to as little as possible. In making corrections in proofs the recognized symbols which will be found in the "Standard Dictionary" should be used. Second (page) proofs will be submitted to authors who should return them promptly.

Illustrations

14. As the *format* of the journals has been standardized, the size adopted being crown quarto (about $7\frac{1}{8}$ in. $\times 9\frac{5}{8}$ in. cut), no text-figure, when printed, should exceed $4\frac{1}{2} \times 5$ inches. Figures for plates should be so planned as to fill a crown quarto plate—the maximum space available for figures being $5\frac{3}{4} \times 8$ inches exclusive of that for letterpress printing.

15. Photos or drawings for illustration should accompany the manuscript and each should bear on the reverse side the name of the paper to which it relates together with the title or legend, figure or plate number, and the size to be reproduced. When giving instructions for reduction linear measurements are understood; thus, "half-size" means reduce to half the length and breadth, not half the area. A photograph should not be rolled up, nor pinned, and should always be packed flat. A complete list of plates and figures should always accompany the paper.

16. Line drawings should be made with clear black lines on smooth white paper, preferably Bristol board. Rough paper should be avoided. Care should

be taken that all the lines are drawn firmly; scratchy or grey lines, produced by the ink being thinned down, are not permissible. Drawings should be larger than the required size. All lettering should be neatly and clearly put in, care being taken to make all lettering sufficiently large to stand reduction.

17. For half-tone work, copy should be made on glossy silver paper and of the same size or larger than the size required.

18. For three-colour work, copy may be oil-painting, water-colour, coloured photograph or coloured transparency, and larger than the size required. In preparing copy, one should use only the primary colours, in any combination, as only inks of primary colours are used in printing. Originals can be enlarged, if necessary, but this should be avoided if possible.

19. For detailed instructions regarding preparation of illustrations, it would be of advantage to refer to Mr. C. M. Hutchinson's article on "Photographic illustrations" in the *Agricultural Journal of India*, Vol. XI, Pt. 3, July 1916, and Mr. A. W. Slater's paper on "The Preparation and Reproduction of Scientific Illustrations" in the *Proceedings of the Third Entomological Meeting*, 1919, which has been reprinted as *Bulletin No. 114 of the Agricultural Research Institute, Pusa.*

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